Available online at www.sciencedirect.com
SCIENCE DIRECT
European Journal of Medicinal Chemistry 41 (2006) 683–696
EUROPEAN JOURNAL OF MEDICINAL CHEMISTRY
http://france.elsevier.com/direct/ejmech

Original paper

Synthesis, stereochemistry, and antimicrobial evaluation of substituted piperidin-4-one oxime ethers

C. Ramalingan [a], Y.T. Park [a,*], S. Kabilan [b]

[a] *Department of Chemistry, Kyungpook National University, Daegu 702-701, Korea*
[b] *Department of Chemistry, Annamalai University, Annamalainagar 608 002, India*

Received in revised form 17 February 2006; accepted 20 February 2006
Available online 04 April 2006

Abstract

In a wide search program toward new and efficient antimicrobial agents, a series of substituted piperidin-4-one oxime ethers (**5a**–**5k**) was synthesized and tested for their in vitro antibacterial and antifungal activities. Also, the structures of these oxime ethers and their relative stereochemistries have been investigated by nuclear magnetic resonance spectroscopy. In all the oxime ethers synthesized, the orientation of the N-O bond of the oxime ether moiety *syn* to C-5 (E-isomer) was deduced based on ^{1}H NMR and ^{13}C NMR spectra. It was found that the sterically less hindered compounds, either C-3 (H) and C-5 (H)- or C-3 (Me) and C-5 (H) -substituted ones **5a**, **5c**, **5d**, **5f**, **5g**, **5i** and **5j** prefer chair conformation, whereas the sterically more hindered C-3 (Me) and C-5 (Me) -substituted ones **5b**, **5e**, **5h,** and **5k** prefer twist-boat conformation. Among the oxime ethers tested, 1,3,5-trimethyl-2,6-diphenylpiperidin-4-one *O*-(2-chlorophenylmethyl)oxime (**5h**) exhibited good antibacterial property against *Bacillus subtilis*, with minimum inhibitory concentration (MIC) closer to that of reference drug, streptomycin. Compounds, 1,3-dimethyl-2,6-diphenylpiperidin-4-one *O*-(2-chlorophenylmethyl)oxime (**5g**) and 1,3-dimethyl-2,6-diphenylpiperidin-4-one *O*-(2-bromophenylmethyl)oxime (**5j**) showed potent antifungal activity against *Aspergillus flavus* and *Candida-51*, respectively. The later compound **5j** is more active than the reference drug while the activity of the former one **5g** is similar to that of the reference drug, amphotericin B in terms of MIC. The present results may be used as key steps for the construction of novel chemical entities with better pharmacological profiles than standard drugs.

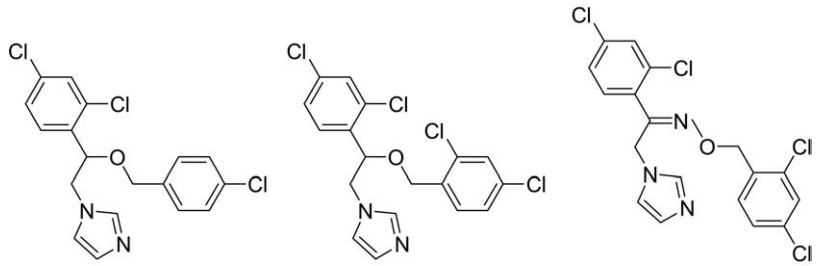
Keywords: Piperidin-4-one oxime ethers; Benzylation; Stereochemistry; Antibacterial activity; Antifungal activity

1. Introduction

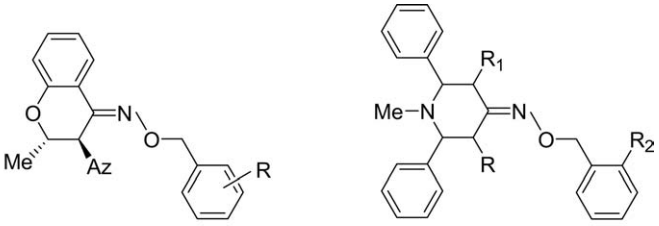
Despite the growing list of antimicrobial agents including polyenes, allyl amines, azole derivatives, fluoropyrimidines, and thiocarbamates, their clinical use has been limited by their relatively high risk of toxicity, insufficiencies in their antimicrobial activity and/or pharmacokinetic deficiencies. These observations foregrounded the emergent need to develop safe, efficacious, and non-toxic antimycotics that can be administered orally as well as parenterally [1–3]. Heterocyclic compounds carrying piperidine skeleton are attractive targets of organic synthesis owing to their pharmacological activity and their wide occurrence in nature [4–6]. Specifically, piperidine-based chemical entities with aryl substituents at carbons 2 and 6 of the piperidine ring have been documented as potent microbial agents [7–15].

* Corresponding author. Tel.: +82 53 950 5334; fax: +82 53 950 6330.
E-mail address: ytpark@knu.ac.kr (Y.T. Park).

0223-5234/$ - see front matter © 2006 Elsevier SAS. All rights reserved.
doi:10.1016/j.ejmech.2006.02.005



Miconazole Econazole Oxiconazole



Azolyl-2-methyl chromanone oxime ethers

Piperidin-4-one oxime ethers (Target chemical entities)

It is known that clinically useful azole antifungal drugs such as miconazole, econazole, and oxiconazole have an *o*-benzyl structural unit in addition to azole and aryl moieties. Particularly, oxiconazole has an oximino unit, which is connected to *o*-benzyl, C-aryl, and C-azole groups. Recently, various oxiconazole-related antifungal chemical entities (ACE), which possess a benzyl unit linked to oximino moiety with different substituents including chromone scaffold have been reported [16,17]. Furthermore, synthesis of novel chemical entities, which are still in resemblance with bioactive molecules by virtue of the presence of some critical structural features, is an essential component of the search for new leads in drug designing programs.

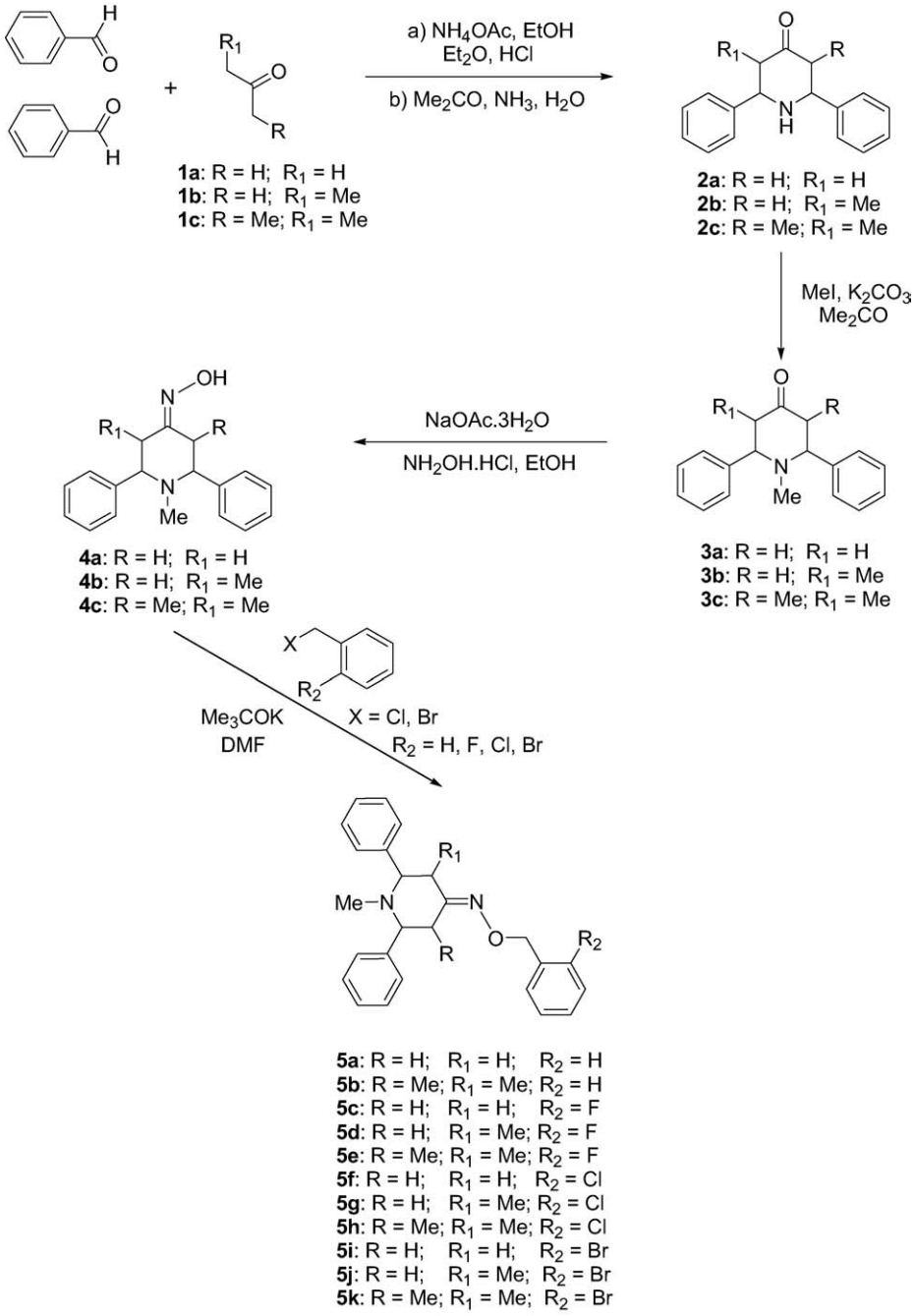
Hence, a careful perusal of literature on antimicrobial agents has induced us to synthesize a class of molecules having 2,6-diarylpiperidin-4-one oxime ethers with various substituents and to evaluate their antimicrobial activity.

2. Results and discussion

2.1. Synthesis

A four step synthetic strategy was adopted for the synthesis of substituted piperidin-4-one oxime ethers (**5a**–**5k**). The general schematic representation describing the routes of syntheses is furnished in Scheme 1. Diphenylpiperidin-4-one **2a** [18,19] was obtained by the condensation reaction of benzaldehyde, acetone (**1a**), and ammonium acetate in 2:1:1 ratio, respectively. The diphenylpiperidin-4-one **2a** afforded 1-methyl-2,6-diphenylpiperidin-4-one (**3a**) upon reflux with iodomethane in the presence of anhydrous potassium carbonate in acetone [20]. Oxime **4a** was obtained by treatment of the 1-methyl-2,6-diphenylpiperidin-4-one (**3a**) with hydroxylamine hydrochloride in the presence of sodium acetate trihydrate in absolute ethanol in good yield [21]. Similarly, the piperidin-4-ones **2b** and **2c** were obtained through the condensation reaction by using 2-butanone (**1b**) and 3-pentanone (**1c**), respectively, with benzaldehyde and ammonium acetate in ethanol. Methylation followed by oximation for **1b** and **1c**, resulted in the corresponding oximes **4b** and **4c,** respectively. All the synthesized substituted piperidin-4-one oximes (**4a**–**4c**) are freely soluble in *N*,*N*-dimethylformamide but insoluble in polar protic solvents such as ethanol, methanol and partially soluble in polar aprotic solvent acetonitrile at room temperature. Formation of ether, 1-methyl-2,6-diphenylpiperidin-4-one *O*-(phenylmethyl) oxime (**5a**) was achieved by benzylation of the oxime, **4a** using potassium tertiary butoxide in *N*,*N*-dimethylformamide at room temperature. The similar methods were adopted for the syntheses of substituted piperidin-4-one oxime ethers (**5b**–**5k**). For the etherification of the oximes, various methods such as sodium in methanol, aqueous potassium hydroxide in dimethylsulfoxide, potassium hydroxide in methanol, potassium carbonate in acetonitrile under refluxed conditions, cesium carbonate in *N*,*N*-dimethylformamide at 0 °C, and sodium hydride in *N*,*N*-dimethylformamide under room temperature were examined. Of these methods, the use of potassium tertiary butoxide in *N*,*N*-dimethylformamide was found to be the proficient method for the etherification (Table 1). The etherification with potassium tertiary butoxide preceded all other methods with excellent yields irrespective of the benzyl halides used (Table 1).

The structure of all the synthesized oxime ethers is established on the basis of nuclear magnetic resonance spectroscopy (^{1}H and ^{13}C NMR), infrared spectrometry (IR), and gas chromatography/mass spectrometry (GC/MS). The reaction yields, physical constants, and results of elemental analyses of the oxime ethers are presented in Table 1. Particularly, the ^{1}H NMR and ^{13}C NMR spectroscopic data clearly furnishes the



Scheme 1.

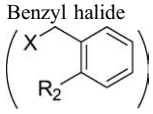
conformations of all the oxime ethers **5a**–**5k** where the N-O bond of the oxime ether moiety directed toward *syn* to the carbon 5. Comparison of the values of chemical shifts for the piperidin-4-one oxime ethers with and without substituents on the carbon 3 clearly reveal their conformations.

2.2. Stereochemistry

For the determination of stereochemistries of the oxime ethers, the ^{1}H NMR spectra of **5j** and **5k** are used as examples (Figs. 1,2 and Table 2). The assignment has been made based on characteristic signal positions of functional groups, spin multiplicity, and comparison with those of parent ketones and structurally similar oxime ethers. Even though there are four and eight enantiomeric pairs of diastereomers are expected for the oxime ethers **5j** and **5k**, respectively, since the heterocyclic rings of **5j** and **5k** have three and four stereogenic centers, respectively, a single set of ^{1}H NMR signals for each of **5j** and **5k** were observed. These observations imply that an enantiomeric pair of the possible diastereomers of the oxime ethers was formed exclusively in each case.

Aromatic proton peaks of three phenyl rings of the oxime ethers, **5j** and **5k** appear in the region of 7.11–7.56 ppm as multiplets. Benzylic proton peaks of oxime ether moieties of

Table 1
Reaction yields, physical properties, and results of elemental analyses of oxime ethers **5a–5k**

Oxime ethers	Oxime	Benzyl halide 	Reaction time (h)	Yield (%)	m.p. (°C)	Elemental analysis C (%) obs. (calc.)	H (%) obs. (calc.)	N (%) obs. (calc.)
5a	**4a**	X = Br; R_2 = H	14	82	78–79	81.11 (81.05)	7.32 (7.07)	7.68 (7.56)
5a	**4a**	X = Cl; R_2 = H	19	80	78–79	–	–	–
5b	**4c**	X = Br; R_2 = H	14	87	67–68	81.09 (81.37)	7.90 (7.59)	7.11 (7.03)
5b	**4c**	X = Cl; R_2 = H	19	90	67–68	–	–	–
5c	**4a**	X = Br; R_2 = F	16	78	76–77	77.25 (77.29)	6.62 (6.49)	7.30 (7.21)
5c	**4a**	X = Cl; R_2 = F	20	75	76–77	–	–	–
5d	**4b**	X = Br; R_2 = F	14.5	84	71–72	77.75 (77.58)	6.85 (6.76)	6.98 (6.96)
5d	**4b**	X = Cl; R_2 = F	20	86	71–72	–	–	–
5e	**4c**	X = Br; R_2 = F	15.5	76	88–89	77.48 (77.76)	7.20 (7.01)	6.75 (6.73)
5e	**4c**	X = Cl; R_2 = F	20	84	88–89	–	–	–
5f	**4a**	X = Br; R_2 = Cl	17	70	80–81	74.31 (74.15)	6.41 (6.22)	7.02 (6.92)
5f	**4a**	X = Cl; R_2 = Cl	24	72	80–81	–	–	–
5g	**4b**	X = Br; R_2 = Cl	17	89	92–93	74.42 (74.54)	6.82 (6.50)	6.75 (6.69)
5g	**4b**	X = Cl; R_2 = Cl	24	93	92–93	–	–	–
5h	**4c**	X = Br; R_2 = Cl	18	91	100–101	75.07 (74.90)	6.97 (6.75)	6.53 (6.47)
5h	**4c**	X = Cl; R_2 = Cl	24	88	100–101	–	–	–
5i	**4a**	X = Br; R_2 = Br	15	76	83–84	66.91 (66.82)	5.76 (5.61)	6.34 (6.23)
5j	**4b**	X = Br; R_2 = Br	15	95	105–106	67.43 (67.39)	6.04 (5.87)	6.16 (6.05)
5k	**4c**	X = Br; R_2 = Br	18	85	93–94	67.75 (67.92)	6.34 (6.12)	5.88 (5.87)

5j and **5k** are observed as two protons singlet in the region of 5.14–5.18 ppm. The methyl protons attached to the piperidine nitrogen of **5j** and **5k** resonate in the region of 1.70–1.76 ppm as singlet with three proton integrals.

For **5j,** one proton peak on carbon 6 appears at 3.18 ppm as a doublet of doublet and one proton peak on carbon 2 at 2.85 ppm as a doublet. The doublet of doublet splitting with coupling constant value of 12.00 and 3.00 Hz at 3.18 ppm and the doublet splitting with coupling constant value of 10.50 Hz at 2.85 ppm are due to two protons of carbon 5 and one proton of carbon 3, respectively. The observations of large values of the vicinal coupling constants between 10.50 and 12.00 Hz and of small value of the vicinal coupling constant of 3.00 Hz for the protons of carbons 6 and 2 of **5j** indicate that the six-member heterocyclic ring of **5j** adopts normal chair conformation with equatorial orientation of phenyl groups at carbons 2 and 6, and equatorial orientation of methyl group at carbon 3. Furthermore, equatorial disposition of phenyl groups at carbons 2 and 6 makes the chair conformation more rigid thereby preventing interconversion from one chair into another. Thus, the relative stereochemistry of one of enantiomeric pair **5j** shown in Fig. 6 is 2S, 3S, 6R. Introduction of a methyl group at equatorial position of carbon 3 shifts the axial proton of carbon 2 resonance towards upfield with a magnitude of around 0.4 ppm (Table 2, see the first column for **5i** and **5j**). This observation is in accord with the result reported by Booth on substituted cyclohexanes [22]. For **5j**, the axial–axial vicinal coupling constant of proton of carbon 2 is around 1.5 Hz less than that of the axial–axial vicinal coupling constant of proton of carbon 6. This difference in coupling constant cannot be due to an electronegativity effect of methyl group since the electronegativity of the methyl group is only slightly different from that of hydrogen. The probable reason is that the heterocyclic ring may be flattened or distorted about the C_2-C_3 bond to decrease gauche interaction between the equatorial phenyl group and the equatorial methyl group at carbons 2 and 3, respectively. A representative ^{1}H NMR spectrum of **5j** is displayed in Fig. 1.

For **5k,** which has two methyl groups at carbons 3 and 5, two doublets at 3.19 and 3.35 ppm with each one proton integral are due to proton of carbons 2 and 6, respectively. Interestingly, abnormal vicinal coupling constants of 3.90 and 9.00 Hz for proton of carbons 2 and 6, respectively, are noticed, compared to those of **5j** (Fig. 2). If the six-member heterocyclic ring of **5k** adopted a normal chair conformation with equatorial orientation of two methyl groups at carbons 3 and 5 along with two equatorial phenyl groups at carbons 2 and 6 akin to **5j**, the observed vicinal coupling constant values of

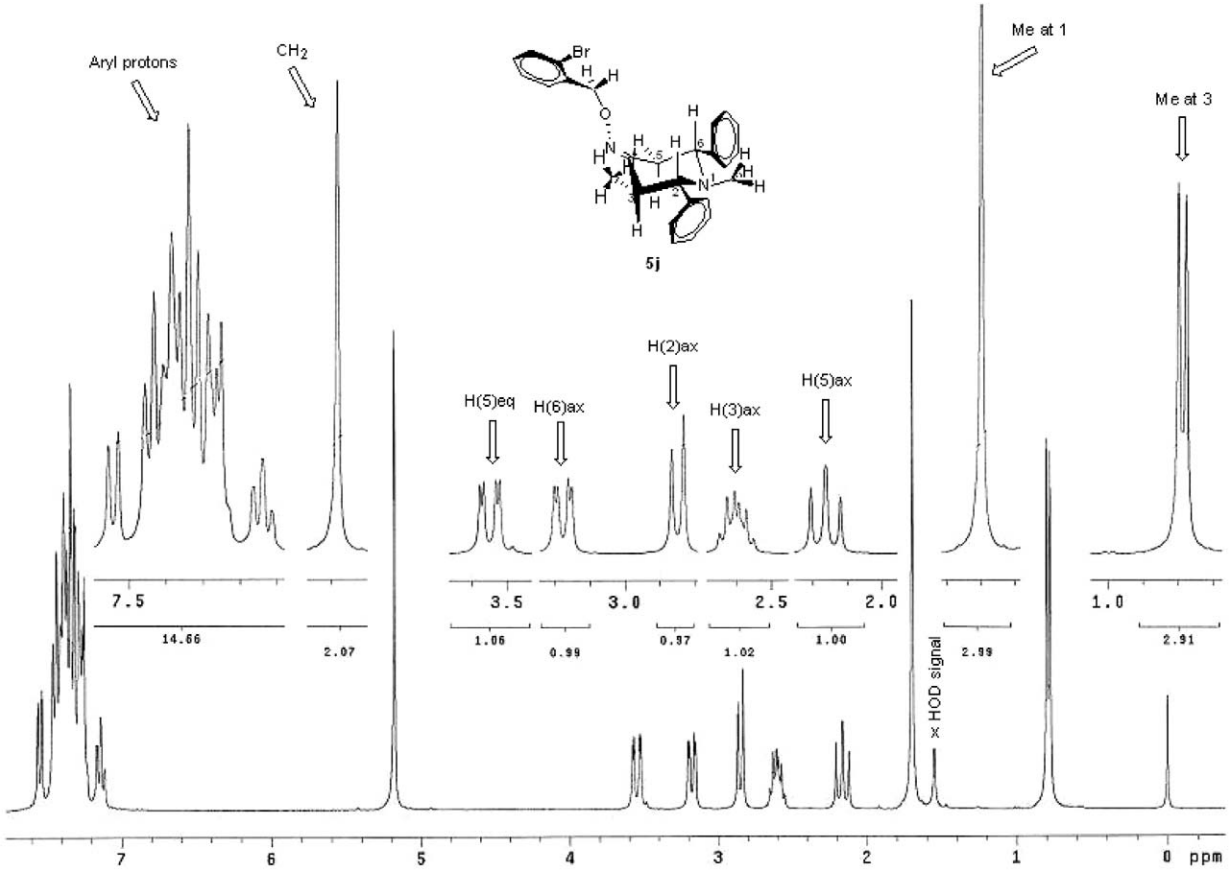
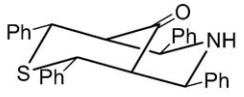


Fig. 1. 300-MHz ^{1}H NMR spectrum of **5j** in $CDCl_3$.

proton of carbons 2 and 6 should be around 10.50 Hz. If the six-member heterocyclic ring of **5k** adopted a rigid boat conformation, the vicinal coupling constant of both the proton of carbons 2 and 6 of **5k** should be less than 5.00 Hz (around 3.00–4.00 Hz) since the coupling value for the vicinal coupling constant in the piperidine ring of thiazine bicyclic compound **6** [23] is known to be 3.90 Hz. The abnormal vicinal coupling constants of proton of carbons 2 and 6 of **5k** indicate that the six-member heterocyclic ring should adopt neither normal chair conformation nor boat conformation (vide infra).



Thiazine bicyclic compound **(6)**

For **5j**, the signal observed at 2.60 ppm with one-proton integrals is corresponding to axial proton of carbon 3 whereas the other one-proton signal at 2.15 ppm is to axial proton of carbon 5. The equatorial proton peak of carbon 5 of **5j** is observed in the unusual downfield region (3.54 ppm). For **5k**, the protons of carbons 3 and 5 absorb at 2.68 and 3.43 ppm, respectively. If this compound adopted a normal chair conformation, both proton peaks would appear near the position where proton peak of carbon 3 of **5j** absorbed. But a slightly deshielding absorption of proton of carbon 3 (~0.1 ppm) is noted, compared to that of **5j**.

Another interesting point here is that since the oxime ethers contain double bond of imino group on **5j**, a free rotation of the imino double bond is restricted. Hence, formation of either geometrical Z or E isomer is expected as ^{1}H NMR provide a single set of signals. If the N-O were *anti* to carbon 5 (or *syn* to carbon 3, Fig. 3), the unusual deshielding absorption of one proton signal (3.54 ppm) should not be observed for the proton on α- carbon to the C=N moiety in the case of **5j**, unlike **5i**. Hence, the N-O bond of the oxime ether moiety takes *syn* configuration to carbon 5 (Fig. 4) rather than *syn* to carbon 3. In other words, an enantiomeric pair of the diastereomers with E-configuration for **5j** has been established.

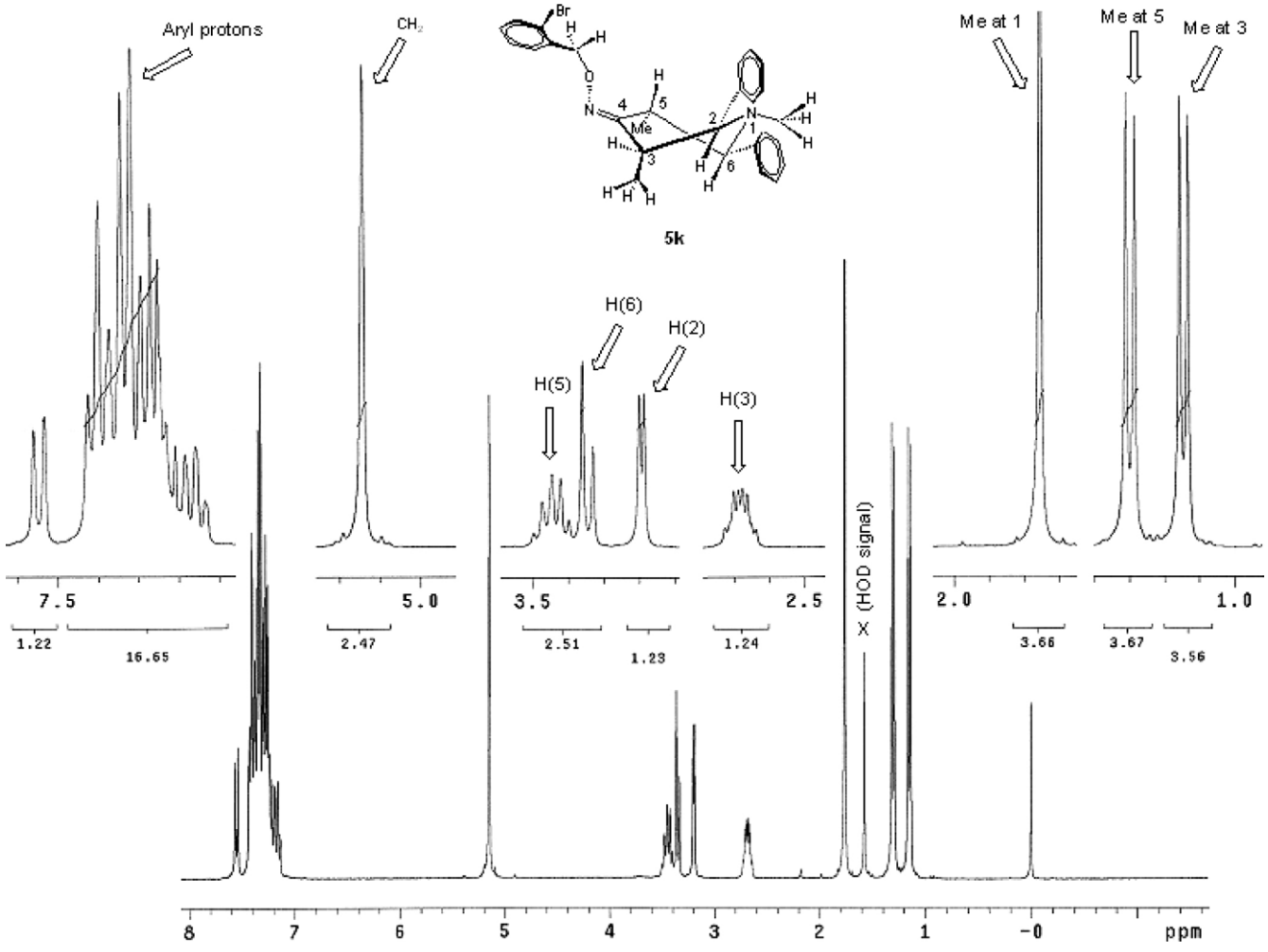


Fig. 2. 300-MHz ^{1}H NMR spectrum of **5k** in $CDCl_3$.

Table 2
^{1}H NMR Chemical shifts of heterocyclic ring protons of oxime ethers **5a–5k**

Compound	H on C(2)	H on C(3)	H on C(5)	H on C(6)
5a	3.24(ax)	2.51(eq)	3.43(eq)	3.17(ax)
		2.48(ax)	2.16(ax)	
5b	3.17	2.67	3.38	3.32
5c	3.24(ax)	2.50(eq)	3.42(eq)	3.15(ax)
		2.48(ax)	2.20(ax)	
5d	2.84(ax)	2.58(ax)	3.48(eq)	3.15(ax)
			2.12(ax)	
5e	3.17	2.67	3.37	3.31
5f	3.25(ax)	2.52(eq)	3.48(eq)	3.19(ax)
		2.49(ax)	2.20(ax)	
5g	2.85(ax)	2.60(ax)	3.54(eq)	3.17(ax)
			2.16(ax)	
5h	3.19	2.68	3.42	3.34
5i	3.25(ax)	2.52(eq)	3.48(eq)	3.19(ax)
		2.49(ax)	2.20(ax)	
5j	2.85(ax)	2.60(ax)	3.54(eq)	3.18(ax)
			2.15(ax)	
5k	3.19	2.68	3.43	3.35

In general, since there are two phenyl groups attached to carbons 2 and 6 with neighboring electronegative nitrogen on **5j** and **5k**, the absorptions of protons of carbons 2 and 6 should

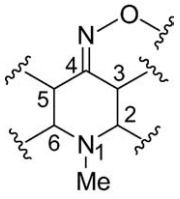


Fig. 3. Configuration of N-O bond *anti* to carbon 5.

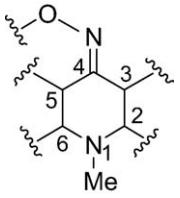
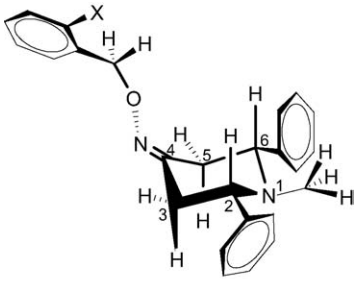


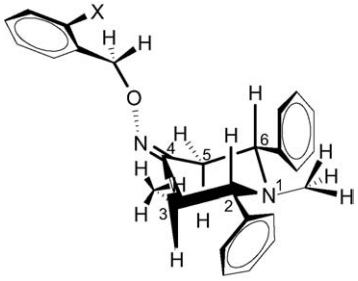
Fig. 4. Configuration of N-O bond *syn* to carbon 5.

be in the downfield region compared to protons of carbons 3 and 5. In contrast, the equatorial proton of carbon 5 is observed in the unusual down field region in these cases as we have seen earlier. The question that remains to be answered is the unusual downfield absorption of equatorial proton of carbon 5. A plausible explanation is that since the N-O bond of the oxime ether



5a; X = H
5c; X = F
5f; X = Cl
5i; X = Br

Fig. 5. Conformation of oxime ethers without methyl substituents at carbons 3 and 5.



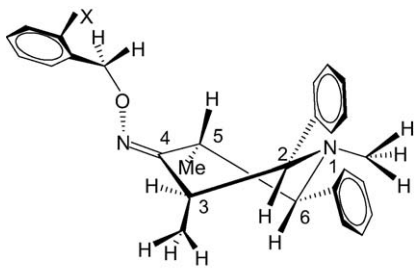
5d; X = F
5g; X = Cl
5j; X = Br

Fig. 6. Conformation of oxime ethers with methyl substituent at carbon 3.

moiety is parallel to the equatorial proton of carbon 5, the oxygen induces the polarity of the bond between carbon 5 and equatorial proton of carbon 5 due to 1,3-spatial proximity effect [24–26]. The equatorial proton of carbon 5 acquires a slight positive and the carbon 5 acquires slight negative charges, and thus, the absorption of equatorial proton of carbon 5 is in the unusual downfield region of **5j.**

Interestingly, the proton of carbon 5 of **5k** is also observed in the unusual downfield region as in the case of **5j**. However, the absorption is comparatively upfield (~0.1 ppm). If the bond between carbon 5 and proton of carbon 5 were perfect axial, then there would not be any spatial interaction with the N-O of the oxime ether moiety and the proton of carbon 5 of this compound. As a result, the proton of carbon 5 should be absorbed in the upfield region compared to the proton of carbon 2 and 6. Hence, the bond between carbon 5 and proton of carbon 5 on **5k** is neither equatorial nor axial.

Moreover, the protons of the methyl group attached to the carbon 3 of **5j** resonate as a doublet at 0.79 ppm with the coupling constant 6.60 Hz, whereas for the protons of the methyl groups attached to the carbons 3 and 5 for **5k** resonate as two doublets in the unusual downfield region of 1.15 and 1.30 ppm with the coupling constants of 7.20 and 7.50 Hz, respectively. The difference in the absorption between the protons of these two methyl groups of **5k** is appreciable (~0.2 ppm). These observations suggest that the six-member heterocyclic ring for **5k**



5b; X = H
5e; X = F
5h; X = Cl
5k; X = Br

Fig. 7. Conformation of oxime ethers with methyl substituents at carbons 3 and 5.

may take a twist-boat conformation unlike **5j** and the relative stereochemistry of one of enantiomeric pair **5k** shown in Fig. 7 is 2R, 3R, 5S, 6S.

Similarly, ^{1}H NMR analyses of **5a–5i** show that the six-member heterocyclic scaffold adopts a chair conformation with the orientation of N-O moiety *syn* to the carbon 5 for **5a**, **5c**, **5d**, **5f**, **5g**, and **5i** whereas the six member heterocyclic ring of **5b**, **5e**, and **5h** adopts a twist-boat conformation similar to **5k**. The relative stereochemistry of each of enantiomers of the diastereomers **5a**, **5c**, **5f** and **5i** shown in Fig. 5 is 2S, 6R akin to that of **5j**. The six-member heterocyclic rings of oxime ethers with C-3 methyl substituent (**5d** and **5g**) and, oxime ethers with C-3 and C-5 two methyl substituents (**5b**, **5e**, and **5h**) have same relative stereochemistry as those of **5j** and **5k**, respectively.

The chemical shifts of ^{13}C NMR of the oxime ethers **5a–5k** and the differences between those of oxime ethers and the corresponding ketones are listed in Tables 3 and 4, respectively. ^{13}C NMR spectra of **5j** and **5k** are used as examples to discuss the conformations of the oxime ethers. Aromatic carbons of **5j** and **5k** are distinguished from other carbons by their characteristic absorption in the region of 127.12–132.53 ppm. Ipso carbons of phenyl group at carbons 2 and 6, and benzyl group resonate between 137.70 and 145.52 ppm whereas carbons, which have bromo substituents, absorb at 122.97 and 122.69 ppm, respectively. The carbons of imino groups of **5j** and **5k** absorb in a downfield of 159.92 and 163.77 ppm, respectively, due to electronegativity of nitrogen and anisotropic effect. The methylenic carbons of benzyl groups of **5j** and **5k** resonate at 74.75 and 74.82 ppm, respectively.

Of the heterocyclic ring carbons for **5j** with 3- methyl group, the peaks of carbons 2 and 6 are observed in the downfield region compared to carbons 3 and 5. It is known that introduction of a methyl group at an equatorial position on the six-member ring makes the nearby carbon shift downward appreciably due to the β-effect of the methyl group [27]. Thus, the peak appearing in the downfield of the two signals (77.74 ppm) is for carbon 2 and the upfield one (69.51 ppm) is for carbon 6. Of the remaining two upfield signals (43.22 and 35.36 ppm) of the ring carbons compared to those of carbons

Table 3
^{13}C NMR Chemical shift values of substituted piperidin-4-one oxime ethers **5a–5k**

Compound	C(2)	C(3)	C(4)	C(5)	C(6)	CH_2 (benzyl)	Me at C(3)	Me at C(5)	Others
5a	70.59	41.20	156.80	35.27	69.23	75.40	–	–	41.20, Me at 1; 143.92, 128.63, 128.30, 128.00, 127.28, aryl carbons
5b	75.77	42.99	163.40	40.63	72.50	75.43	15.92	21.12	42.13, Me at 1; 145.58, 143.37, 138.58, 128.55, 128.37, 128.22, 127.82, 127.46, 127.38, 127.28, 127.03, aryl carbons
5c	70.61	41.20	157.02	35.24	69.27	69.04	–	–	41.20, Me at 1; 144.09, 143.90, 130.36, 129.44, 129.34, 128.63, 127.37, 127.27, 125.05, 123.90, 115.39, 115.10, aryl carbons.
5d	77.73	43.22	159.70	35.24	69.41	69.01	12.58	–	41.55, Me at 1; 144.39, 142.99, 130.59, 129.26, 128.56, 128.39, 128.21, 127.37, 127.20, 123.79, 115.31, 115.04, aryl carbons.
5e	75.77	43.00	163.70	40.60	72.47	69.02	15.90	21.00	42.12, Me at 1; 145.55, 143.30, 130.20, 129.19, 128.53, 128.39, 127.38, 127.25, 127.04, 123.77, 115.26, 114.97, aryl carbons.
5f	70.63	41.20	157.22	35.31	69.32	72.49	–	–	41.20, Me at 1; 144.08, 143.85, 135.83, 129.29, 128.63, 127.37, 127.25, 126.59, aryl carbons.
5g	77.73	43.20	159.85	35.32	69.46	72.54	12.58	–	41.54, Me at 1; 144.37, 142.93, 136.09, 133.17, 129.61, 129.23, 128.60, 128.39, 128.17, 127.37, 127.20, 126.49, aryl carbons.
5h	75.72	42.99	163.72	40.65	72.44	72.60	15.92	21.09	42.15, Me at 1; 145.52, 143.29, 136.26, 132.92, 129.21, 128.53, 128.37, 127.22, 126.52, aryl carbons.
5i	70.70	41.21	157.31	35.36	69.36	74.69	–	–	41.21, Me at 1; 144.08, 137.46, 132.59, 131.46, 129.39, 128.97, 128.66, 127.38, 127.22, 122.85, aryl carbons.
5j	77.74	43.22	159.92	35.36	69.51	74.75	12.59	–	41.55, Me at 1; 144.39, 142.95, 137.70, 132.53, 129.66, 128.87, 128.58, 128.37, 128.19, 127.22, 122.97, aryl carbons.
5k	75.74	42.99	163.77	40.63	72.47	74.82	15.96	21.05	42.13, Me at 1; 145.52, 143.33, 137.91, 132.48, 131.44, 129.34, 128.76, 128.56, 128.40, 127.27, 127.12, 122.69, aryl carbons.

2 and 6, the one in the upfield region (35.36 ppm) is for carbon 5 and the other is for carbon 3. A representative ^{13}C NMR spectrum of **5j** is displayed in Fig. 8. A similar trend is observed for the six-member heterocyclic ring of **5k**. However, an appreciable difference in the magnitude of shielding and deshielding effects is noted for **5k**, compared to those for **5j**.

The chemical shifts of heterocyclic ring carbons of **5j** and **5k** are shielded compared to their values of the parent ketones (**3b** and **3c**) except carbon 2 for **5j**. It is known that a decrease in the electronegativity of a group on a six-member heterocyclic ring shields the α-carbon and deshields the β- and γ- carbons [28]. Since the electronegativity of nitrogen of the C=$NOCH_2Ar$ moiety must be less than that of oxygen of the C=O group, shielding of the α-carbons (carbons 3 and 5) is thus in accordance with the electronegativity effect. Of the two β- carbons (carbons 2 and 6), the *syn* carbon (carbon 6) to the N-O bond of the oxime ether moiety is shielded unlike the other (carbon 2) except **5k**. A comparison between the absorption of carbons 3 and 5 of the oxime ethers (**5j** and **5k**) and their corresponding ketones (**3b** and **3c**) show that the shielding effect for carbon 5, which is *syn* to the N-O bond is higher than that of the *anti* carbon (carbon 3) due to the induced polarity of the equatorial bond of carbon 5 by the electronegative oxygen of the oxime ether moiety (due to 1,3- spatial proximity effect) as discussed in the ^{1}H NMR analysis. The negative charge on the carbon 5 is transmitted to a small extent to the carbon 6 and the shielding thus outweighs the deshielding produced by the electronegativity effect.

Table 4
The difference of ^{13}C NMR chemical shifts of ketones and oxime ethers ($\delta_{ketone}-\delta_{oxime\ ether}$) [a]

Compound	C(2)	C(3)	C(4)	C(5)	C(6)	Me at C(3)	Me at C(4)
5a	–0.70	9.28	49.01	15.21	0.66	–	–
5b	1.79	7.63	45.76	9.99	5.06	–4.80	–10.00
5c	–0.72	9.28	48.79	15.24	0.62	–	–
5d	–0.55	7.66	47.78	15.39	1.00	–1.58	–
5e	1.79	7.62	45.46	10.02	5.09	–4.78	–9.88
5f	–0.74	9.28	48.59	15.17	0.57	–	–
5g	–0.55	7.68	47.63	15.31	0.95	–1.58	–
5h	1.84	7.63	45.44	9.97	5.12	–4.80	9.97
5i	–0.81	9.27	48.50	15.12	0.53	–	–
5j	–0.56	7.66	47.56	15.27	0.90	–1.59	–
5k	1.82	7.63	45.39	9.99	5.09	–4.84	9.93

[a] Positive and negative signs denote shielding and deshielding, respectively.

The deshielding magnitude of the carbons 2 and 3 of oxime ether **5j** compared to those of **5i** is 7.1 and 2.0 ppm, respectively, due to the introduction of an equatorial methyl group at carbon 3. For the corresponding ketones, **3a** and **3b**, these deshielding magnitudes of carbons 2 and 3 are 7.3 and 0.4 ppm, respectively. Hence, α- effect was significantly influenced by adjacent exocyclic polar groups. The absorption of equatorial methyl carbon of **5j** is downward 1.6 ppm compared to its corresponding ketone **3b**. This may probably be due to the less polar effect of the C=N in oxime ethers than that of C=O moiety [29].

For **5k**, a different ^{13}C NMR spectrum is noticed (Fig. 9). Although the difference between deshielding magnitudes of carbon 3 of **5k** and **5j** is small (0.2 ppm), the difference for carbon 2 of **5k** and **5j** is appreciable (~2 ppm). The two methyl carbons of carbons 3 and 5 of **5k** resonate at unusual downfield region (15.96 and 21.05 ppm) and the difference between the absorption of two methyl groups of **5k** is appreciable (5.0–5.2 ppm). Furthermore, the carbon 2 of **5k** absorbs in the appreciable shielding region unlike **5j** compared to the corresponding ketones **3b** and **3c**. If **5k** adopted a normal chair conformation akin to **5j**, the absorption of methyl carbon of carbon 3 should be at around 12.6 ppm since carbon 3 is *anti* to the N-O bond of the oxime ether moiety and, the other methyl group at carbon 5 should ab-

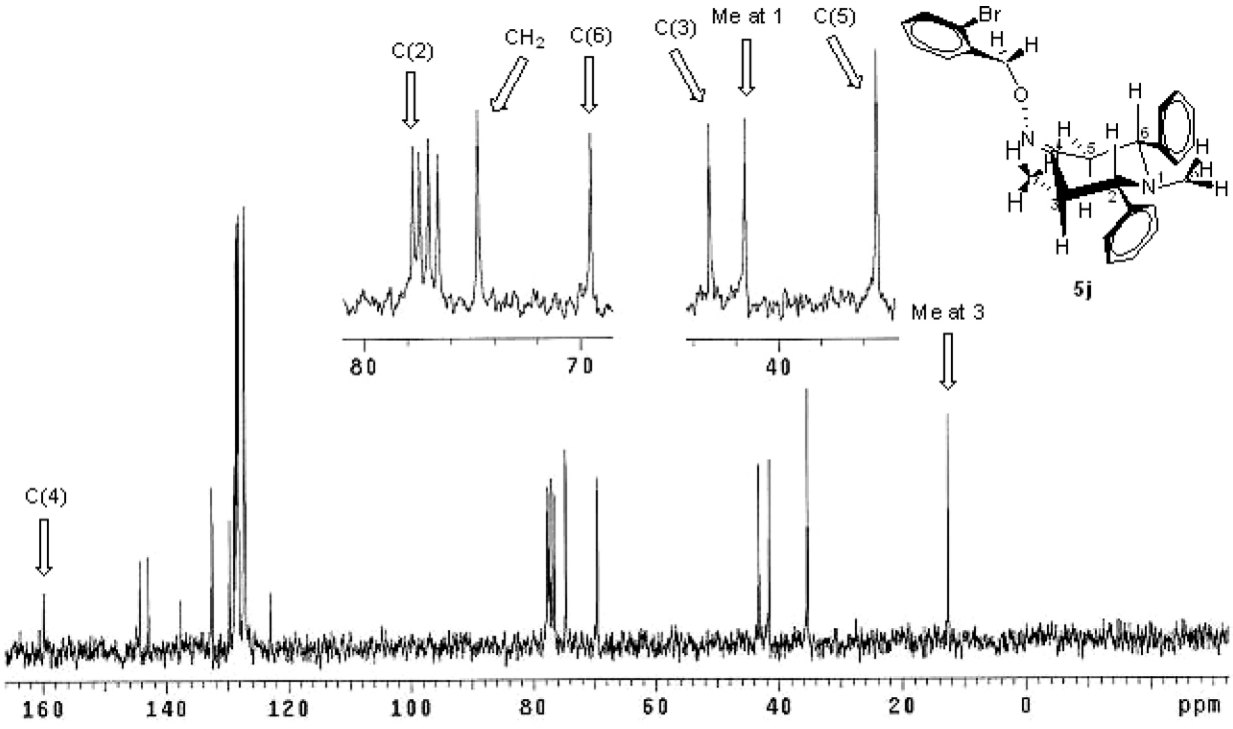


Fig. 8. 75-MHz ^{13}C NMR spectrum of **5j** in $CDCl_3$.

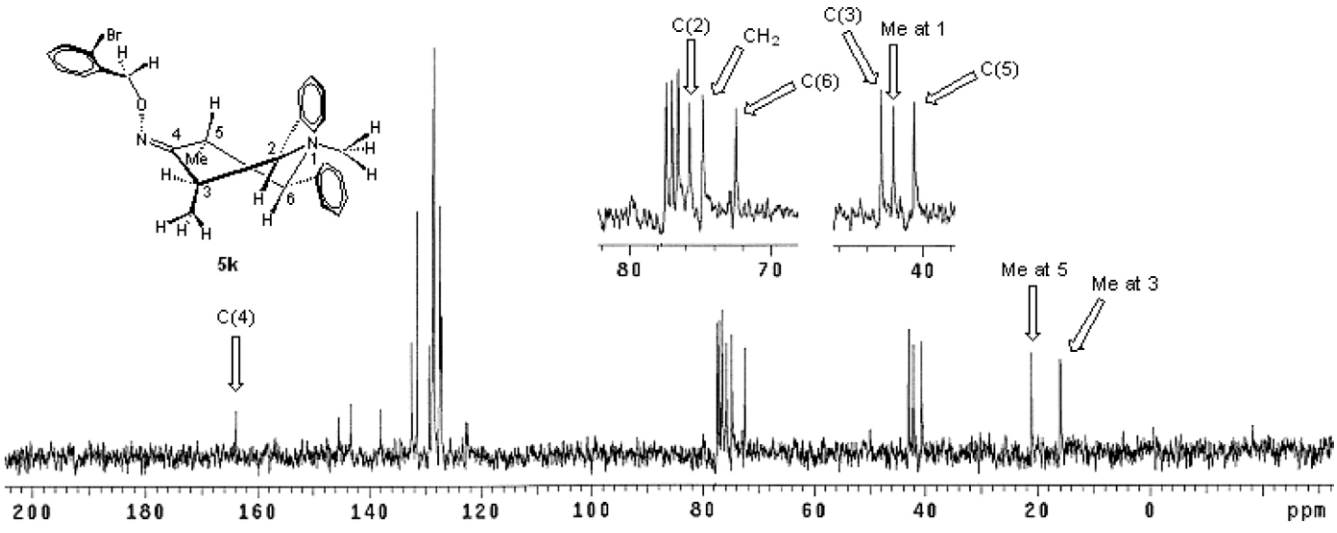


Fig. 9. 75-MHz ^{13}C NMR spectrum of **5k** in $CDCl_3$.

sorb at a still higher field owing to the steric interaction. All these observations suggest that **5k** adopts a twist-boat conformation and particularly the methyl group *syn* to the N-O of the oxime ether moiety avoids steric interaction with the oxime ether group.

2.3. Antimicrobial evaluation

2.3.1. Antibacterial activity

Table 5 shows the in vitro antibacterial activities of the substituted piperidin-4-one oxime ethers **5a**–**5k** and of streptomycin, taken as the reference drug, on a panel of bacterial strains such as *Streptococcus faecalis* (ATCC-29212), *Pseudomonas aeruginosa* (ATCC-27853), *Bacillus subtilis* (ATCC-6033) and *Staphylococcus aureus* (ATCC-25923) as minimum inhibitory concentration (MIC, μg ml^{-1}). Streptomycin was the first antibiotic used against mycobacterium tuberculosis and it was used for years in monotherapy regimens, leading to the emergence of resistance. Besides its other antibiotic properties, it is one of the most widely used agents for dermatological reasons [30,31]. The compounds **5a** and **5b** with and without substituents at carbons 3 and 5 of the six-member heterocyclic moiety along with the benzyl unit did not show in vitro antibacterial activity even at the

Table 5
Antibacterial activity of oxime ethers **5a–5k**

Entry	R	R_1	R_2	MIC, μg ml^{-1}			
				S. faecalis	*P. aeruginosa*	*B. subtilis*	*S. aureus*
5a	H	H	H	> 128	> 128	> 128	> 128
5b	Me	Me	H	> 128	> 128	128	128
5c	H	H	F	> 128	> 128	> 128	> 128
5d	H	Me	F	128	64	> 128	> 128
5e	Me	Me	F	> 128	> 128	> 128	> 128
5f	H	H	Cl	64	64	128	32
5g	H	Me	Cl	64	64	32	32
5h	Me	Me	Cl	32	32	16	32
5i	H	H	Br	64	64	128	> 128
5j	H	Me	Br	64	128	128	64
5k	Me	Me	Br	> 128	64	64	64
Streptomycin				16	16	8	64

Table 6
Antifungal activity of oxime ethers **5a–5k**

Entry	R	R_1	R_2	MIC, μg ml^{-1}			
				A. niger	*Candida-51*	*A. flavus*	*C. albicans*
5a	H	H	H	> 128	> 128	> 128	> 128
5b	Me	Me	H	> 128	> 128	> 128	> 128
5c	H	H	F	128	128	64	128
5d	H	Me	F	128	64	64	64
5e	Me	Me	F	128	128	> 128	> 128
5f	H	H	Cl	64	64	> 128	128
5g	H	Me	Cl	64	32	16	64
5h	Me	Me	Cl	128	> 128	> 128	128
5i	H	H	Br	> 128	32	32	64
5j	H	Me	Br	64	16	128	64
5k	Me	Me	Br	64	128	> 128	128
Amphotericin B				32	16	64	32

maximum concentration of 128 μg ml^{-1} against all the tested organisms except **5b** against *B. subtilis* and *S. aureus*, which exhibited activity at 128 μg ml^{-1}.

Introduction of a fluoro group at *ortho* position of the benzyl moiety along with methyl group at carbon 3 of the six-member heterocyclic ring (**5d**) exerted activity at 64 and 128 μg ml^{-1} against *P. aeruginosa* and *S. faecalis*, respectively, while against *B. subtilis* and *S. aureus*, this compound failed to show activity even at the maximum concentration. Other compounds with fluoro moiety at *ortho* position of the benzyl unit along with and without methyl substituents at carbons 3 and 5 of the heterocyclic ring (**5c** and **5e**) did not show activity even at the maximum concentration against all the tested strains.

Substitution of a chloro group for the fluoro group in **5c**, **5d** and **5e** led to compounds **5f**, **5g** and **5h**, respectively. These three compounds exerted appreciable antibacterial activity at a MIC of 16–64 μg ml^{-1} against all the tested strains except **5f** against *B. subtilis*, which exhibited activity at 128 μg ml^{-1}. Of these, the **5h** was found to be most potent against *B. subtilis*, showing MIC of 16 μg ml^{-1} whereas the activities were 32 μg ml^{-1} for **5f–5h** against *S. aureus*, **5g** against *B. subtilis* and **5h** against *S. faecalis* and *P. aeruginosa*.

Replacement of the chloro group by a bromo group in **5f** (**5i**) did not show appreciable change in the activity against *S. faecalis*, *P. aeruginosa* and *B. subtilis* while the activity was reduced against *S. aureus*. It (**5i**) did not exhibit activity even at the maximum concentration. Instead of chloro functionality, substitution of bromo group in **5g** and **5h** (**5j** and **5k**) showed decrease in activity against all the tested organisms except **5j** against *S. faecalis*, which exerted similar activity as in the case of **5g**.

2.3.2. Antifungal activity

The in vitro antifungal activity of the substituted piperidin-4-one oxime ethers (**5a–5k**) and of Amphotericin B as the reference drug on a panel of fungi such as *Aspergillus niger* (NCIM-590), *Candida-51* (NCIM-C27), *Aspergillus flavus* (NCIM-539) and *Candida albicans* (NCIM-C27), measured as minimum inhibitory concentration (MIC, μg ml^{-1}) is furnished in Table 6. Against all the fungal strains, the **5a** and **5b** without substituents at benzyl unit along with carbons 3 and 5 of the piperidine heterocyclic ring did not exhibit antifungal activity even at the maximum concentration of 128 μg ml^{-1}.

Due to the introduction of fluoro function at *ortho* position of the benzyl unit in **5a** (**5c**), a marked improvement in activity was noticed when compared to **5a** against all the tested strains. Introduction of a methyl group at carbon 3 of the six-member heterocyclic ring in **5c** (**5d**) did not show appreciable change in activity against *A. niger* and *A. flavus* while the activity was enhanced against *Candida-51* and *C. albicans*. Furthermore, substitution of methyl groups at carbons 3 and 5 of the six-member heterocyclic ring in **5c** (**5e**) failed to show activity against *A. flavus* and *C. albicans* whereas against the rest of the organisms tested, it did not affect the activity.

When the fluoro group was replaced by a chloro function in **5d** (**5g**), there was no appreciable change in activity noticed against *C. albicans* while the activity was reduced against *A. flavus*. However, against *A. niger* and *candida-51*, the activity improved. Replacement of hydrogen at carbon 3 of the piperidine ring by a methyl group in **5f** (**5g**) exhibited most potent activity against *A. flavus* whereas against *A. niger*, the activity remained as such as in the case of **5f**. Moreover, replacement of hydrogens at carbons 3 and 5 of the piperidine ring by methyl groups in **5f** (**5h**) failed to exert activity even at the maximum concentration against *Candida-51* and *A. flavus* while the activity was reduced against *A. niger*.

Against *A. niger*, the introduction of bromo function instead of chloro function in **5f** (**5i**) failed to show activity even at the maximum concentration whereas an improvement in activity was noticed by this introduction against the rest of the organisms tested. Methyl substitution at carbon 3 of the piperidine ring in **5i** (**5j**) exhibited most potent activity against *Candida-51*. Against *C. albicans*, this methyl substitution did not change the activity while the activity was reduced against *A. flavus*. However, an appreciable improvement in activity was noticed against *A. niger*. Furthermore, another methyl substitution at carbon 5 of the heterocyclic ring in **5j** (**5k**) did not affect the activity against *A. niger* whereas the activity was reduced against *Candida-51*, *A. flavus* and *C. albicans*.

3. Conclusion

Several substituted piperidin-4-one oxime ethers were synthesized starting with benzaldehyde and respective ketones through the pathway involving Mannich reaction, methylation, oximation, and etherification. The nuclear magnetic resonance studies clearly furnish the conformations of the oxime ethers along with the orientation of N-O bond *syn* to carbon 5. The chair conformation of the six-member heterocyclic ring carrying a methyl substituent on carbon 3 and without substituents on carbons 3 and 5 of the oxime ethers and the twist-boat conformation of the six-member heterocyclic ring carrying a methyl substituent on each of carbons 3 and 5 of the oxime ethers were deduced based on NMR studies. The microbiological study was undertaken to evaluate the effects on the antibacterial and antifungal activities of the substituted piperidin-4-one oxime ethers. The data shown in Tables 5 and 6 indicate that the oxime ethers, which contain chloro functions/bromo functions at the *ortho* position of the benzyl unit with and without methyl substituents at carbon 3/carbons 3 and 5 of the six-member heterocyclic scaffold, play an important role in eliciting biological response. Particularly, of the oxime ethers tested, the compound with chloro substituent at the *ortho* position of the benzyl moiety along with the methyl groups at carbons 3 and 5 of the six-member heterocyclic ring exhibited potent activity against the bacterial strain, *Bacillus subtilis*. The oxime ethers with methyl substituent at carbon 3 of the piperidine heterocyclic ring along with chloro/bromo substituents at the *ortho* position of the benzyl moiety exerted potent antifungal activity against the fungal strains, *Aspergillus flavus* and *Candida-51*, respectively. A careful observation of the results implies that the nature of the halogen substituent on the *ortho* position of the benzyl moiety along with twist-boat conformation of the heterocyclic ring carrying methyl substituents at carbons 3 and 5 is determinant towards antibacterial activity whereas towards antifungal activity, the nature of the halogen substituent on the ortho position of the benzyl unit along with classical chair conformation of the piperidine ring carrying methyl group at carbon 3 is determinant. However, the mode of action of these oxime ethers is unknown. These observations may promote further development of our research in this field. Further development of this group of oxime ethers may lead to chemical entities with better pharmacological profile than standard drugs. Thus, in the future this class of oxime ethers may be used as templates for the construction of better drugs to combat bacterial and fungal infections.

4. Experimental section

4.1. Chemistry

4.1.1. General

All solvents were distilled prior to use. All reagents were reagent grade and were used without further purification. Thin layer chromatography was performed on Silica gel 60 F plates eluting with the solvents indicated. Column chromatography was performed on Silica gel 230–400 mesh slurry packed in glass columns (2 cm × 20 cm) with eluent as indicated. All the reported melting points were measured in open capillaries and are uncorrected. ^{1}H NMR and ^{13}C NMR spectra were acquired at 300 and 75 MHz, respectively, by using $CDCl_3$ as a solvent for all the compounds except **4c** and TMS as an internal standard. For **4c,** C_5D_5N was used as a solvent due to the poor solubility in $CDCl_3$. The abbreviations s, d, dd and m stand for the resonance multiplicities singlet, doublet, doublet of doublet and multiplet, respectively.

The parent 2,6-diarylpiperidin-4-ones (**2a**–**2c**) were synthesized through Mannich reaction by adopting literature method [18,19].

4.1.2. Synthesis of 1-methyl-2,6-diphenylpiperidin-4-one *(3a)*

In a 50 ml round-bottomed flask, fitted with a reflux condenser, were placed 2,6-diphenylpiperidin-4-one (5.02 g, 0.02 mol), anhydrous potassium carbonate (3 g), acetone (30 ml), and iodomethane (1.56 ml, 0.025 mol). The mixture in the flask was refluxed at 45–55 °C for 3.5 h. After cooling, the solvent along with excess methyl iodide was stripped off from the reaction mixture under reduced pressure. Addition of distilled water (200 ml) followed by ammonia (5 ml) yielded a crude solid. The crude solid thus obtained was filtered, washed with distilled water. Crystallization from ethanol yielded the title compound (4.8 g, 91%). Similar procedure was adopted for the synthesis of 1,3-dimthyl-2,6-diphenylpiperidin-4-one (**3b**) and 1,3,5-trimthyl-2,6-diphenylpiperidin-4-one (**3c**). The physical constants of all the synthesized compounds agree well with the values in the literature [20].

4.1.3. Synthesis of 1-methyl-2,6-diphenylpiperidin-4-one oxime ***(4a)***

To the ethanolic solution of 1-methyl-2,6-diphenylpiperidin-4-one (5.3 g, 0.02 mol, 60 ml) in a two neck round-bottomed flask fitted with a reflux condenser, was added sodium acetate trihydrate (8.16 g, 0.06 mol). The mixture was heated to boil for 15 min. Then, hydroxylamine hydrochloride (2.07 g, 0.03 mol) was added to the above mixture and was heated to reflux for further 1.5 h. After cooling, when the mixture was slowly poured into ice-cold water (400 ml) with constant stirring, the crude product was precipitated. Crystallization from absolute ethanol yielded the pure title compound. (5.0 g, 90%). m.p.: 186–187 °C (m.p. 188 °C [21]). The above method was adopted for the synthesis of 1,3-dimethyl-2,6-diphenylpiperidin-4-one oxime (**4b**, m.p.: 212–213 °C, m.p. 220 °C [21]) and 1,3,5-trimethyl-2,6-diphenylpiperidin-4-one oxime (**4c**, m.p.: 193–195 °C, m.p. 194–196 °C [21]).

4.1.3.1. Typical method of synthesis of 1-methyl-2,6-diphenylpiperidin-4-one O-(phenylmethyl)oxime ***(5a)***. To a stirring solution of 1-methyl-2,6-diphenylpiperidin-4-one oxime (0.28 g, 1 mmol) in *N*,*N*-dimethylformamide (16 ml) at normal room temperature, was added potassium *tert*-butoxide (0.12 g, 1 mmol). After stirring for 25 min, benzyl bromide (0.11 ml, 0.95 mmol) was added in drop-wise fashion to the reaction mixture and the stirring was continued for further 14 h at room temperature. Then, the content of the flask was quenched with brine (50 ml) and diluted with diethyl ether (30 ml). The organic layer was separated and the aqueous layer was extracted with diethyl ether (2 × 30 ml). The combined organic layers were dried over magnesium carbonate, filtered and concentrated under reduced pressure. Purification of the oxime ether was accomplished by column chromatography over silica gel, eluting with a solvent system of *n*-hexane: ethyl acetate (40:3). The fractions containing product were collected and concentrated under reduced pressure to yield a white solid: m.p. 78 °C; IR (KBr) (cm^{-1}): 2934, 2908, 2782 (C–H stretching), 1652 (C–N stretching), 1601, 1496 (C=C ring stretching), 759 (N–O stretching); ^1H NMR (δ ppm): 7.45–7.24 (m, 15H) aryl protons, 5.08 (s, 2H) benzyl CH_2, 3.43 (d, $J = 14.10$ Hz, 1H) $H_{5(eq)}$, 3.24 (1H) $H_{2(ax)}$, 3.17 (dd, $J = 12.00$ Hz; 3.30 Hz, 1H) $H_{6(ax)}$, 2.51–2.48 (m, 2H) $H_{2(ax)}$ and $H_{3(ax)}$, 2.16 (1H) $H_{5(ax)}$, 1.76 (s, 3H) CH_3 at 1-position. GC-MS (*m*/*z*): 370 (M^+), (M.F: $C_{25}H_{26}N_2O$), 91 (100%).

The 2,6-diphenylpiperidin-4-one oxime ethers **(5b–5k)** were synthesized from their corresponding oximes in a manner as described for **(5a)**.

4.1.3.2. 1,3,5-Trimethyl-2,6-diphenylpiperidin-4-one O-(phenylmethyl)oxime ***(5b)***. IR (KBr) (cm^{-1}): 2961, 2924, 2841, 2789 (C–H stretching), 1632 (C=N stretching), 1601, 1495 (C=C ring stretching), 749 (N–O stretching). ^1H NMR (δ ppm): 7.39–7.20 (m, 15H) aryl protons, 5.07 (s, 2H) benzyl CH_2, 3.41–3.36 (m, 1H) H_5, 3.32 (d, $J = 9.00$ Hz, 1H) H_6, 3.17 (d, $J = 3.90$ Hz, 1H) H_2, 2.69–2.65 (m, 1H) H_3, 1.74 (s, 3H) CH_3 at 1, 1.29 (d, $J = 7.50$ Hz, 3H) CH_3 at 5, 1.10 (d, $J = 6.60$ Hz, 3H) CH_3 at 3. GC-MS (*m*/*z*): 398 (M^+), (M.F: $C_{27}H_{30}N_2O$), 91 (100%).

4.1.3.3. 1-Methyl-2,6-diphenylpiperidin-4-one O-(2-fluorophenylmethyl)oxime ***(5c)***. IR (KBr) (cm^{-1}): 2995, 2967, 2794 (C–H stretching), 1651 (C=N stretching), 1601, 1491 (C=C ring stretching), etc. ^1H NMR (δ ppm): 7.45–7.01(m, 14H) aryl protons, 5.15 (s, 2H) benzyl CH_2, 3.42 (dd, $J = 14.10$ Hz; 1.80 Hz, 1H) $H_{5(eq)}$, 3.24 (1H) $H_{2(ax)}$, 3.15 (dd, $J = 12.00$ Hz; 3.30 Hz, 1H) $H_{6(ax)}$, 2.50–2.47 (m, 2H) $H_{2(ax)}$ and $H_{3(ax)}$, 2.20 (1H) $H_{5(ax)}$, 1.77 (s, 3H) CH_3 at 1. GC-MS (*m*/*z*): 388 (100%, M^+), (M.F: $C_{25}H_{25}FN_2O$).

4.1.3.4. 1,3-Dimethyl-2,6-diphenylpiperidin-4-one O-(2-fluorophenylmethyl)oxime ***(5d)***. IR (KBr) (cm^{-1}) 2994, 2976, 2937, 2777 (C–H stretching), 1642 (C=N stretching), 1600, 1492 (C=C ring stretching), 756 (N–O stretching). ^1H NMR (δ ppm): 7.44–7.01 (m, 14H) aryl protons, 5.16 (s, 2H) benzyl CH_2, 3.48 (dd, $J = 13.80$ Hz; 3.30 Hz, 1H) $H_{5(eq)}$, 3.15 (dd, $J = 11.85$ Hz; 3.15 Hz, 1H) $H_{6(ax)}$, 2.84 (d, $J = 10.20$ Hz, 1H) $H_{2(ax)}$, 2.61–2.56 (m, 1H) $H_{3(ax)}$, 2.12 (1H) $H_{5(ax)}$, 1.69 (s, 3H) CH_3 at 1, 0.79 (d, $J = 6.60$ Hz, 3H) CH_3 at 3. GC–MS (*m*/*z*): 402 (100%, M^+), (M.F: $C_{26}H_{27}FN_2O$).

4.1.3.5. 1,3,5-Trimethyl-2,6-diphenylpiperidin-4-one O-(2-fluorophenylmethyl)oxime ***(5e)***. IR (KBr) (cm^{-1}): 2968, 2933, 2873, 2791 (C–H stretching), 1623 (C=N stretching), 1600, 1491 (C=C ring stretching), 748 (N–O stretching). ^1H NMR (δ ppm): 7.41–7.01 (m, 14H) aryl protons, 5.14 (s, 2H) benzyl CH_2, 3.40–3.35 (m, 1H) H_5, 3.31 (d, $J = 9.00$ Hz, 1H) H_6, 3.17 (d, $J = 3.90$ Hz, 1H) H_2, 2.69–2.65 (m, 1H) H_3, 1.74 (s, 3H) CH_3 at 1, 1.28 (d, $J = 7.20$ Hz, 3H) CH_3 at 5, 1.10 (d, $J = 6.60$ Hz, 3H) CH_3 at 3. GC–MS (*m*/*z*): 416 (M^+), (M.F: $C_{27}H_{29}FN_2O$), 120 (100%).

4.1.3.6. 1-Methyl-2,6-diphenylpiperidin-4-one O-(2-chlorophenylmethyl)oxime ***(5f)***. IR (KBr) (cm^{-1}): 2995, 2967, 2905, 2793 (C–H stretching), 1647 (C=N stretching), 1595, 1494 (C=C ring stretching), 756 (N–O stretching). ^1H NMR (δ ppm): 7.47–7.19 (m, 14H) aryl protons, 5.19 (s, 2H) benzyl CH_2, 3.48 (dd, $J = 14.10$ Hz; 2.40 Hz, 1H) $H_{5(eq)}$, 3.25 (1H) $H_{2(ax)}$, 3.19 (dd, $J = 12.00$ Hz; 3.60 Hz, 1H) $H_{6(ax)}$, 2.52–2.48 (m, 2H) $H_{2(ax)}$ and $H_{3(ax)}$, 2.20 (1H) $H_{5(ax)}$, 1.77 (s, 3H) CH_3 at 1. GC-MS (*m*/*z*): 404 (100%, M^+), (M.F: $C_{25}H_{25}ClN_2O$).

4.1.3.7. 1,3-Dimethyl-2,6-diphenylpiperidin-4-one O-(2-chlorophenylmethyl)oxime ***(5g)***. IR (KBr) (cm^{-1}): 2995, 2967, 2782 (C–H stretching), 1641 (C=N stretching), 1601, 1492 (C=C ring stretching), 753 (N–O stretching). ^1H NMR (δ ppm): 7.45–7.18 (m, 14H) aryl protons, 5.21 (s, 2H) benzyl CH_2, 3.54 (dd, $J = 13.80$ Hz; 3.30 Hz, 1H) $H_{5(eq)}$, 3.17 (dd, $J = 11.85$ Hz; 3.15 Hz, 1H) $H_{6(ax)}$, 2.85 (d, $J = 10.20$ Hz, 1H) $H_{2(ax)}$, 2.63–2.56 (m, 1H) $H_{3(ax)}$, 2.16 (1H) $H_{5(ax)}$, 1.70 (s, 3H) CH_3 at 1, 0.79 (d, $J = 6.60$ Hz, 3H) CH_3 at 3. GC–MS (*m*/*z*): 418 (M^+), (M.F: $C_{26}H_{27}ClN_2O$), 264 (100%).

*4.1.3.8. 1,3,5-Trimethyl-2,6-diphenylpiperidin-4-one O-(2-chlorophenylmethyl)oxime **(5h)***. IR (KBr) (cm^{-1}): 2975, 2965, 2936, 2789 (C–H stretching), 1638 (C=N stretching), 1599, 1492 (C=C ring stretching), 752 (N–O stretching). ^1H NMR (δ ppm): 7.42–7.19 (m, 14H) aryl protons, 5.18 (s, 2H) benzyl CH_2, 3.46–3.39 (m, 1H) H_5, 3.34 (d, $J = 9.00$ Hz, 1H) H_6, 3.19 (d, $J = 3.90$ Hz, 1H) H_2, 2.70 – 2.66 (m, 1H) H_3, 1.75 (s, 3H) CH_3 at 1, 1.30 (d, $J = 7.50$ Hz, 3H) CH_3 at 5, 1.14 (d, $J = 6.90$ Hz, 3H) CH_3 at 3. GC–MS (*m*/*z*): 432 (M^+), (M.F: $C_{27}H_{29}ClN_2O$), 278 (100%).

*4.1.3.9. 1-Methyl-2,6-diphenylpiperidin-4-one O-(2-bromophenylmethyl)oxime **(5i)***. IR (KBr) (cm^{-1}): 2966, 2944, 2905, 2847, 2792 (C–H stretching), 1646 (C=N stretching), 1594, 1493 (C=C ring stretching), 755 (N–O stretching). ^1H NMR (δ ppm): 7.55–7.11 (m, 14H) aryl protons, 5.16 (s, 2H) benzyl CH_2, 3.48 (d, $J = 14.40$ Hz, 1H) $H_{5(eq)}$, 3.25 (1H) $H_{2(ax)}$, 3.19 (dd, $J = 11.85$ Hz; 3.15 Hz, 1H) $H_{6(ax)}$, 2.52–2.49 (m, 2H) $H_{2(ax)}$ and $H_{3(ax)}$, 2.20 (1H) $H_{5(ax)}$, 1.78 (s, 3H) CH_3 at 1. GC-MS (*m*/*z*): 448 (M^+-1), (M.F: $C_{25}H_{25}BrN_2O$), 207 (100%).

*4.1.3.10. 1,3-Dimethyl-2,6-diphenylpiperidin-4-one O-(2-bromophenylmethyl)oxime **(5j)***. IR (KBr) (cm^{-1}): 2995, 2967, 2901, 2782 (C–H stretching), 1641 (C=N stretching), 1601, 1493 (C=C ring stretching), 751 (N–O stretching). ^1H NMR (δ ppm): 7.56–7.11 (m, 14H) aryl protons, 5.18 (s, 2H) benzyl CH_2, 3.54 (dd, $J = 13.80$ Hz; 3.30 Hz, 1H) $H_{5(eq)}$, 3.18 (dd, $J = 12.00$ Hz; 3.00 Hz, 1H) $H_{6(ax)}$, 2.85 (d, $J = 10.50$ Hz, 1H) $H_{2(ax)}$, 2.63–2.57 (m, 1H) $H_{3(ax)}$, 2.16 (1H) $H_{5(ax)}$, 1.70 (s, 3H) CH_3 at 1, 0.79 (d, $J = 6.60$ Hz, 3H) CH_3 at 3. GC–MS (*m*/*z*): 462 (M^+-1), (M.F: $C_{26}H_{27}BrN_2O$), 120 (100%).

*4.1.3.11. 1,3,5-Trimethyl-2,6-diphenylpiperidin-4-one O-(2-bromophenylmethyl)oxime **(5k)***. IR (KBr) (cm^{-1}): 2964, 2930, 2838, 2785 (C–H stretching), 1634 C=N stretching), 1593, 1492 (C=C ring stretching), 752 (N–O stretching). ^1H NMR (δ ppm): 7.56–7.14 (m, 14H) aryl protons, 5.14 (s, 2H) benzyl CH_2, 3.50–3.40 (m, 1H) H_5, 3.35 (d, $J = 9.00$ Hz, 1H) H_6, 3.19 (d, $J = 3.90$ Hz, 1H) H_2, 2.73–2.64 (m, 1H) H_3, 1.76 (s, 3H) CH_3 at 1, 1.30 (d, $J = 7.50$ Hz, 3H) CH_3 at 5, 1.15 (d, $J = 7.20$ Hz, 3H) CH_3 at 3. GC–MS (*m*/*z*): 476 (M^+-1), (M.F: $C_{27}H_{29}BrN_2O$), 207 (100%).

4.2. Microbiological evaluation

The in vitro antibacterial and antifungal activities were performed by the twofold serial dilution technique. The media used for the bacteria and fungi were Nutrient broth (NB) and Sabourauds dextrose broth (SDB), respectively. Microdilution panels were prepared containing twofold dilutions of the compounds and standard drugs in dimethyl sulfoxide (DMSO) ranging from 8 to 128 μg ml^{-1}. Seeded broth (broth containing microbial spores) was prepared in NB from 24-h-old bacterial cultures on nutrient agar at 37 ± 1 °C while fungal spores from 24- to 168-h-old Sabourauds agar slant cultures were suspended in SDB. The colony forming units (cfu) of the seeded broth were determined by plating method and adjusted in the range of 10^2–10^5 cfu ml^{-1}. Testing was performed at pH 7.4 ± 0.2. A set of assay tubes containing only inoculated broth was kept as control and likewise solvent controls were also run simultaneously. The tubes were incubated in BOD incubators at ~37 °C for bacteria and ~28 °C for fungi. The minimum inhibitory concentrations (MICs) were recorded by visual observations after 24 h (for bacteria) and 72–96 h (for fungi) of incubation. For the standard, streptomycin (for bacteria) and amphotericin B (for fungi) were used while DMSO was used as solvent control.

Acknowledgements

This work was supported by Korea Research Foundation Grant (KRF-2004-005-C00006).

References

[1] D.J. Sheehan, C.A. Hitchcock, C.M. Sibley, Clin. Microbiol. Rev. 12 (1999) 40–79.
[2] G.P. DeMuri, M.K. Hostetter, Pediatr. Clin. North Am. 42 (3) (1995) 665–685.
[3] M.A. Ghannoum, L.B. Rice, Clin. Microbiol. Rev. 12 (4) (1999) 501–517.
[4] Atta-ur-Rahman, in: S.R. Angle, J.G. Breitenbucher (Eds.), Stereoselective Synthesis, Elsevier, New York, 1995, pp. 453–502.
[5] S.W. Pelletier, M.J. Schneider (Eds.), Chemical and Biological Perspectives, Wiley, New York, 1996, pp. 155–355.
[6] A.R. Kartritzky, W.Q. Fan, J. Org. Chem. 55 (1990) 3205–3209 (and references cited therein).
[7] S. Balasubramanian, C. Ramalingan, G. Aridoss, S. Kabilan, Eur. J. Med. Chem. 40 (7) (2005) 694–700.
[8] S. Balasubramanian, C. Ramalingan, G. Aridoss, P. Parthiban, S. Kabilan, Med. Chem. Res. 13 (5) (2004) 297–311.
[9] C. Ramalingan, S. Balasubramanian, S. Kabilan, M. Vasudevan, Eur. J. Med. Chem. 39 (6) (2004) 527–533.
[10] C. Ramalingan, S. Balasubramanian, S. Kabilan, M. Vasudevan, Med. Chem. Res. 12 (1) (2003) 41–55.
[11] C. Ramalingan, S. Balasubramanian, S. Kabilan, M. Vasudevan, Med. Chem. Res. 12 (1) (2003) 26–40.
[12] I.G. Mobio, A.T. Soldatenkov, V.O. Fedorov, E.A. Ageev, N.D. Sergeeva, S. Lin, E.E. Stashenko, N.S. Prostakov, E.I. Andreeva, Khim. Farm. Zh. 23 (4) (1989) 421–427.
[13] T.K. Mandal, I.G. Mobio, V.V. Kuznetsov, A. Litvinov Zh, E.N. Denisov, V.O. Fedorov, E.I. Andreeva, A.T. Soldatenkov, N.S. Prostakov, Khim. Farm. Zh. 25 (6) (1991) 28–33.
[14] N. Ramesh kumar, A. Veena, R. Ilavarasan, M. Adiraj, P. Shanmugapandiyan, S. Sridhar, Biol. Pharm. Bull. 26 (2) (2003) 188–193.
[15] A.T. Soldatenkov, A.N. Levov, I.G. Mobio, E.V. Polyakova, S.V. Kutyakov, A.L. Tuan, A.I. Komarova, K.B. Polyanskii, E.I. Andreeva, L.I. Minaev, Khim. Farm. Zh. 37 (10) (2003) 526–528.
[16] A. Rosello, S. Bertini, A. Lapucci, M. Macchia, A. Martinelli, S. Rapposelli, E. Herreros, B. Macchia, J. Med. Chem. 45 (22) (2002) 4903–4912.
[17] S. Emami, M. Falahati, A. Banifatemi, A. Shafiee, Bioorg. Med. Chem. Lett. 12 (2004) 5881–5889.
[18] V. Baliah, A. Ekambaram, T.S. Govindarajan, Curr. Sci. 23 (1954) 264.
[19] C.R. Noller, V. Baliah, J. Am. Chem. Soc. 70 (1948) 3853–3855.
[20] M. Balasubramanian, N. Padma, Tetrahedron 19 (1963) 2135–2143.
[21] M. Uma, K. Ragothaman, Indian J. Chem 19B (1) (1980) 74–76 (and references cited therein).
[22] H. Booth, H. Tetrahedron 22 (2) (1966) 615–620.
[23] K. Pandiarajan, K.K. Stalin, J. Indian, Chem. 24B (5) (1985) 565–567.
[24] A.C. Cope, R.J. Cotter, G.G. Roller, J. Am. Chem. Soc. 77 (1955) 3590–3594.

[25] K. Pandiarajan, A. Manimekalai, J. Indian, Chem. 43B (12) (2004) 2696–2699.
[26] K. Pandiarajan, A. Manimekalai, G. Rajarajan, J. Indian, Chem. 39B (7) (2000) 517–524.
[27] D.K. Dalling, D.M. Grant, J. Am. Chem. Soc. 89 (1967) 6612–6622.
[28] J.B. Lambert, D.A. Netzel, H.N. Sun, K.K. Lilianstrom, J. Am. Chem. Soc. 98 (1976) 3778–3783.
[29] F.J. Weigert, J.D. Roberts, J. Am. Chem. Soc. 92 (1970) 1347–1350.
[30] R. Le Gall, C. Marchand, J. Rees, Eur. J. Derm. 15 (3) (2005) 146–151.
[31] P. Ruiz, F. Rodriguez-Cano, F.J. Zerolo, M. Casal, Microbial Drug Resistance 8 (2) (2002) 147–149 (and references cited therein).