Canadian Chemical Transactions Year 2015 | Volume 3 | Issue 3 | Page 261-274

Research Article

DOI:10.13179/canchemtrans.2015.03.03.0205

Synthesis, Spectral characterization of some 3-alkyl-2,6-di(naphthalen-1-yl)piperidin-4-one and oxime derivatives and their screening for Antimicrobial properties

Mariadoss Arockia doss, Sambandam Savithiri, Sandhirakasu Vembu, Govindasamy Rajarajan*and Venugopal Thanikachalam

Department of Chemistry, Annamalai University, Annamalainagar-608002, Tamilnadu, India.

.

Corresponding Author: Email: rajarajang70@gmail.com Tel: +919444147388

Received: *May 16, 2015* ***Revised:*** *June 17, 2015* ***Accepted:*** *June 21, 2015* ***Published:*** *June 23, 2015*

Abstract: Several 3-alkyl-2,6-di(naphthalen-1-yl)piperidin-4-ones were synthesised by the condensation of 1-naphthaldehyde, appropriate ketones and ammonium acetate in ethanol (**1-4**) and their oximes have been synthesised by the direct condensation of the corresponding 3-alkyl-2,6-di(naphthalen-1-yl)piperidin-4-one with hydroxylamine hydrochloride (**5,6**). The synthesised compounds have been characterised by elemental analyses, FT-IR, ^{1}H NMR and ^{13}C NMR spectra and for compound **2** HOMOCOSY and HMBC spectra have also been recorded. The observed chemical shifts and coupling constants suggested that compounds **1-6** adopt normal chair conformation with equatorial orientation of all substituents. The effect of oximation on ring carbons, their associated protons, alkyl substituents and *ipso* carbons are studied. All the synthesised compounds were screened for their antibacterial activity against *S.typhi*, *E.coli, B.subtilis, S.aureus* and *Pseudomonas* and antifungal activity against *A.niger, C.albicans, A.flavus*, *Rhizopus sp, Mucor* and they exhibited better activities against all the tested bacterial and fungal strains.

Keywords: FT-IR, ^{1}H NMR, ^{13}C NMR, antibacterial activity, antifungal activity.

1. INTRODUCTION

Heterocyclic compounds bearing piperidine skeleton are interesting targets of organic synthesis owing to their pharmacological activity and their wide existence in nature [1-3]. Piperidine is a very important pharmocophore because of its presence in numerous alkaloids, pharmaceuticals, agrochemicals and synthetic intermediates. Biologically active alkaloids of substituted piperidine ring systems have been targeted for their total or partial synthesis [4]. Nitrogen containing heterocyclic compounds especially piperidine-4-ones presumably gaining considerable importance owing to varied biological properties such as antibacterial [5], antifungal [6], antiviral [7], anti-tumour [8], analgesic [9], anti-inflammatory, local anesthetic [10], CNS and depressant activities [11]. Earlier studies have been carried out by substitution in 2, 3, 6 positions and evaluate the resulting compounds for their biological activity. It was proposed therefore, that this synthesis of a new series of substituted 4-piperidones would result in compounds with

potent biological activity [12]. Further blocking of its keto group have also shown to exhibit antibacterial [13,14] and antifungal [15] and anticonvulsant activities [16]. The synthesis of piperidine is easy, economic and less time consuming. The parent molecule is flexible in nature and hence various derivatives can be easily prepared by altering its substituents [17]. NMR spectroscopy is a versatile tool for the structural diagnosis of most of the heterocyclic compounds [18-20]. Vicinal proton- proton coupling constants have been widely used in deriving information regarding the conformation of 2r,6c-diarylpiperidin-4-ones [21,22] and their oximes [23,24]. The present study is an attempt to synthesize various piperidine-4-one oxime derivatives and evaluate their antimicrobial activity.

2. EXPERIMENTAL SECTION

2.1 Materials and methods

All the solvents used were of spectral grade. The melting points of all the compounds were measured in open capillaries and are uncorrected. IR spectra were recorded on an AVATAR-330 FT-IR spectrometer (Thermo Nicolet) using KBr (pellet form). ^{1}H NMR spectra were recorded at 500 MHz on Bruker, spectrometer using DMSO-d_6 as solvent for all the compounds. Tetramethylsilane (TMS) was used as an internal reference for all NMR spectra; ^{13}C NMR spectra were recorded at 125 MHz spectrometer in DMSO-d_6. For recording ^{1}H NMR spectra solutions were prepared by dissolving about 10 mg of the compound in 0.5mL of DMSO-d_6. While for recording ^{13}C NMR spectra, about 50 mg of the compound was dissolved in the same volume of solvent. The chemical shifts are reported in δ units (parts per million) relative to the standard. ^{1}H NMR splitting patterns are designated as singlet (s), doublet (d), doublet of doublet (dd), triplet (t), quartet (q) and multiplet (m). Coupling constants are expressed in Hertz (Hz). Sample was prepared by dissolving about 1 mg in 5mL of methanol. Micro analyser was performed on VarioMicro V2.2.0 CHN analyser. Microbial screening studies were made at Pondicherry centre for biological sciences in Pondicherry.

2.2 Synthesis

2.2.1 Synthesis of 3-alkyl-2,6-di(naphthalen-1-yl)piperidin-4-ones (1-5)

The compounds were synthesized following the procedure described for the preparation of 2,6-diarylpiperidin-4-ones by Noller et al. [25], Colapert et al. [26], Rastogi et al.[27]. A mixture of ammonium acetate (0.06 mol), 1-naphthaldehyde (0.12 mol) and appropriate ketone (0.06 mol) in distilled ethanol was heated to boiling. After cooling, the viscous liquid obtained was dissolved in ether (300 ml) and shaken with 15 mL of concentrated hydrochloric acid. The precipitated hydrochloride of 3-alkyl-2,6-di(naphthalene-1-yl)piperidin-4-one was removed by filtration and washed first with a mixture of ethanol and ether (1:1) and then with ether to remove most of the coloured impurities. The base was liberated from an alcoholic solution by adding aqueous ammonia and then diluted with water. It was recrystallized from alcohol.

3-Methyl-2r,6c–di(naphthalene-1-yl) piperidin-4-one (1)

Yield 80%., m.p. 194 (ºC), MF: $C_{26}H_{23}NO$: IR (KBr) (cm^{-1}): 3466 (N-H stretching), 3111-3046 (-CH-Ar stretching), 2982-2928 (-CH-Aliphatic stretching), 1706 (C=O stretching), 1455 (C=C stretching), 777 (Aromatic C-H out of plane bending vibration), 740 (Aromatic C-C out of plane bending vibration).

3, 3-dimethyl-2r,6c–di(naphthalene-1-yl) piperidin-4-one (2)

Yield 85%., m.p. 198 (ºC), MF: $C_{27}H_{25}NO$: IR (KBr) (cm^{-1}): 3474 (N-H stretching), 3106-3049 (-CH-Ar stretching), 2927-2854 (-CH-Aliphatic stretching), 1707 (C=O stretching), 1448 (C=C stretching), 776 (Aromatic C-H out of plane bending vibration), 738 (Aromatic C-C out of plane bending vibration).

3-ethyl-2r,6c–di(naphthalene-1-yl) piperidin-4-one (3)

Yield 75%., m.p. 232 (ºC), MF: $C_{27}H_{25}NO$: IR (KBr) (cm^{-1}): 3490 (N-H stretching), 3096-2993 (-CH-Ar stretching), 2926-2856 (-CH-Aliphatic stretching), 1706 (C=O stretching), 1444 (C=C stretching), 775 (Aromatic C-H out of plane bending vibration), 746 (Aromatic C-C out of plane bending vibration).

3-pentyl-2r,6c–di(naphthalene-1-yl) piperidin-4-one (4)

Yield 80%., m.p. 243ºC), MF: $C_{30}H_{31}NO$: IR (KBr) (cm^{-1}): 3379 (N-H stretching), 3128-3051 (-CH-Ar stretching), 2953-2862 (-CH-Aliphatic stretching), 1708 (C=O stretching), 1516 (C=C stretching), 775 (Aromatic C-H out of plane bending vibration), 735 (Aromatic C-C out of plane bending vibration).

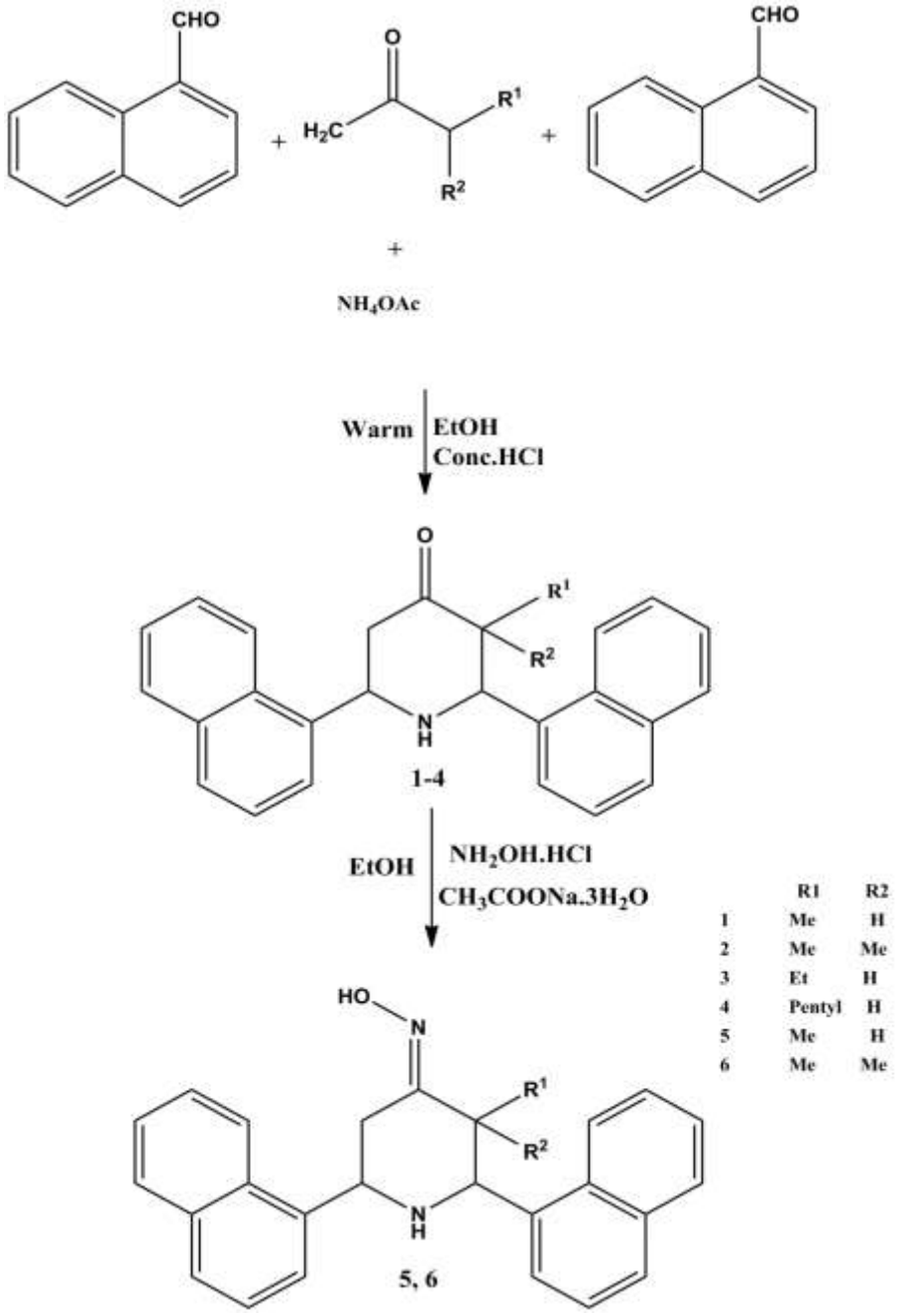

Figure 1. Synthetic route for 3-alkyl-2, 6–di(naphthalene-1-yl)peridin-4-ones (1-4) and their oxime derivatives (5,6).

2.2.2 Synthesis of 3-alkyl-2,6-di(naphthalen-1-yl)piperidin-4-one oxime (5,6)

About 0.05 mol of 3-alkyl-2,6-di(naphthalen-1-yl)piperidin-4-one and sodium acetate trihydrate (0.15 mol) were dissolved in boiling ethanol and hydroxylamine hydrochloride (0.06 mol) was added. The mixture was heated to 40°C and stirred for 3-4 h. It was allowed to stand overnight and then poured into crushed ice. The separated solid oxime was filtered off and recrystallized from ethanol.

3-methyl-2r,6c–di(naphthalene-1-yl) piperidin-4-one oxime (5)

Yield 72%., m.p.238 (ºC), MF: $C_{26}H_{24}N_2O$: IR (KBr) (cm^{-1}): 3240 (N-H stretching), 3300 (OH stretching), 3150-3041 (-CH-Ar stretching), 2965-2849 (-CH-Aliphatic stretching), 1591 (C=N stretching), 1433 (C=C stretching), 730 (Aromatic C-H out of plane bending vibration), 648 (Aromatic C-C out of plane bending vibration).

3,3-dimethyl-2r,6c–di(naphthalene-1-yl) piperidin-4-one oxime (6)

Yield 70%., m.p. 241 (ºC), MF: $C_{27}H_{26}N_2O$: IR (KBr) (cm^{-1}): 3270 (N-H stretching), 3304 (OH stretching), 3145-3058 (-CH-Ar stretching), 2965-2854 (-CH-Aliphatic stretching), 1592 (C=N stretching), 1437 (C=C stretching), 733 (Aromatic C-H out of plane bending vibration), 651 (Aromatic C-C out of plane bending vibration).

2.3. Antimicrobial studies

2.3.1. Antibacterial activity by disc diffusion method

Nutrient agar plates were prepared under sterile conditions and incubated overnight to detect contamination. About 0.2 mL of working stock culture was transferred into separate nutrient agar plates and spread thoroughly using a glass spreader. Whatmann No.1 discs (6 mm in diameter) were impregnated with the test compounds dissolved in DMSO (200 mg/ml) for about half an hour. Commercially available drug disc *(Ciprofloxacin* 10 µg/disc) was used as positive reference standard. Negative controls were also prepared by impregnating the disc of same size in DMSO solvent. The discs were placed on the inoculated agar plates and incubated at 37 ± 1°C for about 18-24 h. Antibacterial activity was evaluated by measuring the zone of inhibition against the test organisms.

2.3.2. Antifungal activity by disc diffusion method

Sabouraud's dextrose agar (SDA) medium was used for the growth of fungi and testing was done in Sabouraud's dextrose broth (SDB) medium. The subculture and the viable count were carried out by the same procedure used for antibacterial studies except the temperature, which should be maintained at 28 ± 1°C for about 72 h. Similarly for disc diffusion method, the petridishes were incubated at 28 ± 1°C for about 72 h. The same concentration of the test compound, solvent (DMSO) and *Amphotericin B* (standard) prepared previously were used for the antifungal studies.

2.3.3 Minimum Inhibitory Concentration (MIC)

The lowest concentration of the test compounds which caused apparently the inhibition of growth of organism was taken as the minimum inhibitory concentration. The MIC was recorded by visual observation after 24 h (bacteria) and 72-96 h (fungi) of incubation. The sterile distilled water and DMSO did not show any inhibition.

3. RESULTS AND DISCUSSION

3.1 Elemental analysis

The compounds **1-4** were synthesized by the condensation of 1-naphthaldehyde, appropriate ketone and ammonium acetate. The compounds **5, 6** were also synthesized by the direct condensation of the corresponding piperidin-4-one with hydroxylamine hydrochloride in the presence of sodium acetate trihydrate (**Figure 1**). All the synthesized compounds are obtained in good yield. All the experimental

values are in good agreement with calculated values. The elemental analyses provide further support to the assignment of structure. The analytical data are given in Table 1.

The compounds **1-6**, ^{1}H and ^{13}C spectra have been recorded and HOMOCOSY and HMBC spectra have also been recorded for compound **2**. The chemical shifts values are given in Tables 2- 8.

Table 1. Analytical data of compounds (1-6).

Compd	m.p (°C)	Yield(%)	Molecular Formula	Elemental analysis (%)			Molecular weight
				C	H	N	
				Found (Calcd)	Found (Calcd)	Found (Calcd)	
1	194	80	$C_{26}H_{23}NO$	85.39 (85.45)	6.31 (6.34)	3.80 (3.83)	365.47
2	232	85	$C_{27}H_{25}NO$	85.30 (85.45)	6.60 (6.64)	3.65 (3.69)	379.49
3	198	75	$C_{27}H_{25}NO$	85.42 (85.45)	6.63 (6.64)	3.65 (3.69)	379.49
4	243	80	$C_{30}H_{31}NO$	85.45 (85.47)	7.39 (7.41)	3.30 (3.32)	421.57
5	238	72	$C_{26}H_{24}N_2O$	82.05 (82.07)	6.35 (6.36)	7.34 (7.36)	380.48
6	241	70	$C_{27}H_{26}N_2O$	82.19 (82.20)	6.64 (6.64)	7.08 (7.10)	394.51

3.2 NMR Spectra

3.2.1. ^{1}H NMR

The two triplets at 6.25 and 6.03 ppm are observed with the coupling constant of J_{aa} = 11.0 Hz and 10.5 Hz, respectively, which may be due to the benzylic protons H_{2a} and H_{6a}. The signals at 3.90 and 3.46 ppm are due to the axial protons H_{3a} and H_{5a}. The lower frequency signal, i.e. 2.88 ppm is assigned to H_{5e}. The upfield triplet at 0.80 ppm is due to methyl protons at C_3. The signals appeared in the region of 7.61 – 8.54 ppm with 14 protons integral are due to naphthyl protons at C-2 and C-6. Compounds **2**, **3** and **4** also have similar kind of splitting like that of **1**. The various ^{1}H chemical shifts of compounds **1-6** are given in Table 2.

The axial and equatorial methylene protons at C-5 [H_{5a} and H_{5e}] are observed at 2.08 and 3.77 ppm, respectively. The methine proton appears at 2.73 ppm. The upfield signal at 0.81 ppm is due to the methyl protons at C-3. The strong downfield signal at 10.71 ppm (integral corresponding to one proton) is due to hydroxyl proton of the oxime group. The proton chemical shift, in this case H-2, resonates at lower frequency relative to H-6. This is due to the magnetic anisotropic effect of the methyl group at C-3. The chemical shifts of the other heterocyclic ring protons are in the order $H_{5e} > H_{3a} > H_{5a}$.

The chemical shifts of 3-alkyl-2,6-di(naphthyl)piperidine-4-one oximes **5** and **6** have been compared with those of the corresponding 3- alkyl-2,6-di(naphthyl)piperidine-4-one **1** and **2**. Conversion

of piperidone to oxime shields all the protons except H_{5e}. The shielding magnitude observed on H_{5e} can be explained.

In compounds **5** and **6** there can be severe interaction between N-O and *syn* equatorial α(C-H) bond, the *syn*-α equatorial α(C-H) bond is said to be polarized and *syn* α –equatorial hydrogen gets a slight positive charge and the *syn* α- carbon acquires a slight negative charge. The negative charge on the syn α- carbon C-5 is transmitted to *syn* α- axial hydrogen to some extent, consequently axial hydrogen at C-5 is shielded while equatorial hydrogen is deshielded.

Table 2. ^{1}H chemical shifts (ppm) of compounds (1-6).

Compound	H(2)	H(3)	H(5a)	H(5e)	H(6a)	-CH_2	-CH_3	Aromatic protons
1	6.25	3.90	3.46	2.88	6.03	–	0.80	7.61-8.54
2	5.15	–	2.57	3.02	5.08	–	1.25, 0.78	7.50-8.38
3	6.27	3.89	2.87	3.89	6.06	1.24,1.48	0.69	7.19-8.77
4	6.26	3.90	2.87	3.90	6.05	0.85-1.52	0.62	7.61-8.80
5	4.77	2.73	2.08	3.77	4.69	-	0.81	7.46-8.37
6	4.92	-	2.17	3.68	4.72	-	0.88, 1.26	7.49-8.38

3.2.2. ^{13}C NMR

In the ^{13}C NMR spectrum of compound **1,** the aromatic carbons could be readily distinguished by their characteristic absorption around125 ppm. The *ipso* carbon should absorb downfield (~135 ppm) compared to other aromatic carbons. Assignments for the heterocyclic ring carbons and naphthylic carbons have been made on the basis of known effects of alkyl substituents in six membered ring compounds. In compound **1**, the signals for the heterocyclic ring carbons appeared at 58.81, 48.22, 204.23, 45.59 and 55.05 ppm. The most downfield signal at 204.23 ppm is obviously due to carbonyl carbon *i.e.*, C-4. The upfield signal at 10.59 ppm is due to methyl carbon at C-3. Among the remaining signals, the downfield signal at 58.81 ppm is assigned to C-2 carbon based on the known deshielding β-effect of the methyl group at C-3 in compound **1**. The other downfield signal at 55.05 ppm is obviously due to C-6. The signal at 48.22 ppm is assigned to C-3 based on known α-effect of the methyl group at C-3. The signal at 45.59 ppm is due to C-5. The ^{13}C NMR chemical shift values of compounds **2** and **4**, which show similar kind of splitting and are assigned like that of compound **1**. In compound **5,** the signals for the heterocyclic ring carbons appeared at 79.65, 56.37, 158.57, 44.06 and 61.88 ppm. The most downfield signal at 158.57 ppm is obviously due to C=N *i.e.* C-4. The upfield signal at 56.37 ppm is due to C-5. Among the remaining signals, the downfield signal at 79.65 ppm is assigned to C-2 carbon based on the known deshielding α-effect of the methyl group. The other downfield signal at 61.88 ppm is due to C-6. The remaining signal at 44.06 ppm is assigned to C-3 based on known α-effect of methyl group at C-5. The compound **6**, exhibits similar type of splitting likes that of compound **5**.The ^{13}C chemical shifts of compounds **1-6** are given in Table 3.

Table 3. ^{13}C chemical shifts (ppm) of compounds (1-6)

Compound	C (2)	C (3)	C (4)	C (5)	C (6)	CH_2	-	$-CH_3$	Aromatic carbons
1	58.81	54.22	204.23	42.59	45.05		–	10.59	123.10-133.71
2	61.42	60.73	212.39	40.56	46.71		–	21.78, 20.84	123.39-139.91
3	57.33	53.54	204.07	42.97	44.85	28.25	1	11.97	123.24-133.77
4	57.11	51.89	203.63	42.44	44.37	31.44-30.91	2	13.48	122.70-133.22
5	58.69	56.37	158.57	44.06	41.88		-	12.77	123.59-140.29
6	57.91	57.07	161.85	42.71	43.36		-	11.26, 14.33	123.28-140.78

Table 4. Chemical shift differences between the α-carbons ($\Delta\delta_\alpha$) in Oximes (**6-10**) and ketones (**1-5**).

Oxime	Piperidone	$\delta_{anti} - \delta_{syn}$ (ppm)		
		Oxime	Piperidone	Oxime - Piperidone
5	1	12.31	2.63	9.68
6	2	14.36	10.17	4.19

Table 5. ^{1}H chemical shift differences (ppm) between phenyl piperidone and its oximes and naphthalene piperidone and its oximes ($\delta_{\text{piperidone and its oximes}} - \delta_{\text{Naphthalene piperidone and its oximes}}$)[a].

Compound	H2	H3	H5e	H5a	H6
1	-2.62	-	-0.72	-0.05	-1.93
2	-1.33	-	0.36	-0.54	-1.02
3	-2.54	-	-0.14	-1.28	-1.99
4	-1.53	-1.25	-0.14	-1.25	-1.99
5	-1.22	-0.17	-0.03	-0.12	-0.78
6	-1.15	-	-0.12	0.06	-0.84

- Could not be determined accurately

a- positive values indicating shielding and negative values deshielding.

It is seen from Table 4 that the conversion of 1 and 2 to the corresponding oximes 5 and 6 shield all the ring carbons except C-2 and methylene carbon of alkyl group at C-3 where slight deshielding has been observed. It is well established that increase in the electronegativity of a group in the ring skeleton deshields α-carbon but shields β and γ carbons. The electronegativity of oxime group should be less than that of C=O group since C=N bond is less polar than C=O bond. Based on the electronegativity effect, one can expect shielding of α -carbons (C-3 and C-5) and deshielding of β-carbons C-2 due to oxime formation. The observed downfield resonance of *anti* β-carbon (C-2) in **5** and **6** relative to the 3-alkylpiperidones **1** and **2** are in accordance with this expectation. The upfield resonances of C-4 in **5** (158.57 ppm) and **6** (161.85 ppm) compared to the corresponding piperidones **1** (204.23 ppm) and **2** (212.39 ppm) are also in accordance with lower electronegativity of the oxime group compared to the carbonyl group. The magnitude of shielding observed on C-5 *i.e.*, *syn* α-carbon (12.77, 11.26 ppm) due to oxime formation is greater than that observed on C-3 *i.e.*, *anti* α-carbon (10.99, 8.02 ppm), similar effects were observed by Pandiarajan *et al*. [28]. Oximation shields H-2 and H-6 protons and methine proton H-3 slightly and there is no appreciable change in the chemical shift of methyl protons of alkyl group at C-3. Comparison of ^{1}H and ^{13}C chemical shifts of 3-alkyl-2,6-bis(1-naphthyl) piperidin-4-ones **1-4** and **5-6** with 3-alkyl-2,6-diphenylpiperidin-4-one [29-31] reveal that all the hetercyclic ring protons are deshielded (~2 ppm) and the heterocyclic ring carbons are shielded(~10ppm). The deshielding and shielding magnitudes are only due to incorporation of the naphthyl group at C-2 and C-6 and this is due to magnetic anisotropic effect. The ^{1}H and ^{13}C chemical shift differences between phenyl piperidone and its oxime and naphthyl piperidone and its oxime are shown in Tables 5 and 6, respectively.

3.3 2D NMR

In the HOMOCOSY spectrum of compound **2**, the signal at 3.02 ppm has cross peaks with the signals at 2.57 and 5.08 ppm. Hence, the signal at 3.02 ppm is due to axial proton at C-5. The signal at 2.57 ppm has cross peak only with the signal at 3.02 ppm. Therefore, the signals at 3.02 and 2.57 ppm are, respectively, due to H_{5a} and H_{5e}. The signal at 5.08 ppm has cross peak only with the signal at 3.02 ppm. Hence, the H-6 signal at 5.08 ppm is due to the proton at C-6. Since the signals for H-6, H-5 and H-2 appear as poorly resolved signals, the *vicinal* coupling and *geminal* coupling constants could not be extracted from these signals. Therefore, it is difficult to study the conformation of compound **2** using *vicinal* and *geminal* coupling constants. HOMOCOSY correlation is shown in Table 7.

Generally, in the HMBC spectrum of compound **2**, the cross peaks are obtained between carbon and hydrogen separated by two or three bonds. The one bond coupling between C and H may be absent or appear as straddled peak. The assignments of ^{1}H NMR and ^{13}C NMR data are also confirmed by the HMBC spectral data (Table 8)**.** In the HMBC spectrum of compound **2**, the signal at 20.84 ppm has cross peaks with protons at 5.15 and 0.78 ppm. The signals at 5.15 and 0.78 ppm are assigned to H-2 and C-3 methyl protons, respectively. Hence, the peak at 20.84 ppm should be due to one of the methyl carbons.

The peak at 21.78 ppm shows cross peak only with the signal at 1.26 ppm. The signal at 1.26 ppm is due to methyl protons of another group at C-3. Hence, the signal at 21.78 ppm should be due to another methyl carbon. It is interesting to note that the signal at 21.78 ppm has straddled peak with the signal at 0.78 ppm which is due to the same methyl protons. The peak at 50.73 ppm shows cross peaks with the signals at 1.26 and 0.78 ppm and are assigned to the two kinds of methyl protons at C-3. Hence, the signal at 50.73 ppm should be due to C-3.The peak at 56.71 ppm shows cross peaks with the signals at 5.15, 5.08 and 3.02 ppm and are assigned to H-6 naphtyl ring proton at C-6 and H-5a, respectively. Hence, the signal at 56.71 ppm should be due to C-6.The peak at 61.42 ppm shows cross peaks with the signals at 5.15, 1.26 and 0.78 ppm. The signal at 5.15, 1.26 and 0.78 ppm are assigned to naphthyl ring proton H-3a

and methyl protons at C-3, respectively. Hence, the peak at 61.42 ppm should be due to C-2. It is also interesting to note that the signal at 61.42 ppm has straddled peak with the signal at 5.15 ppm which is due to H-2. The peak at 212 ppm shows cross peaks with the signals at 1.26 and 0.78 ppm, which is already assigned to two methyl groups at C-3. Hence, the signal at 212 ppm should be due to C-4.

Table 6. ^{13}C chemical shift differences (ppm) between phenyl piperidone and its oximes and naphthalene piperidone and its oximes (1-6) ($\delta_{\text{piperidone and its oximes}}$-$\delta_{\text{Naphthalene piperidone and its oximes}}$)[a].

Compound	C2	C3	C4	C5	C6
1	9.59	3.38	5.27	5.31	6.45
2	8.08	-0.93	0.31	6.64	4.89
3	9.27	-	4.23	-	-
4	10.13	5.24	5.7	6.15	7.51
5	-9.49	-13.17	1.83	-10.26	-1.08
6	2.59	-15.37	2.25	-12.61	-2.26

a - positive values indicating shielding and negative values deshielding.

Table 7. ^{1}H-^{1}H COSY correlation of compound 2.

^{1}H chemical shift		^{1}H-^{1}H COSY correlation
3.02	(H-5a)	2.57 (H-5e), 5.08 (H-6)
2.57	(H-5e)	3.02 (H-5a)
5.08	(H-6)	3.02 (H-5a)

3.4 Antibacterial Evolution (Structure-Activity Relationship)

Antibacterial activity

In vitro antibacterial activity studies of the various 3-alkyl substituted-2,6-bis(1-napthyl) piperidine-4-ones **1-4** and their oximes **5-6** against a group of bacterial strains such as *S.typhi, E.coli, B.subtilis, S.aureus* and *Pseudomonas* have recorded a minimum inhibitory concentration (MIC/mL) and are reproduced in Table 9 it shows that all the compounds exhibit a varied range 6.25-200 µg/mL of antibacterial potency against the tested bacterial strains. Compound **1**, was inactive against *B. subtilis, E.coli* and *pseudomonas*, but active against *S.aureus* and *S.typhi*.

The introduction of another methyl group at C-3 position of **1**(compound **2**) exhibited excellent activity against all the tested bacterial strains. But replacement of methyl group by ethyl group at C-3 position **1**(compound **3**) showed better activity against all the tested bacterial strains except *S.typhi*. Compound **4,** substitution of bulkier pentyl group at C-3 position instead of methyl group, has registered excellent activity against *Pseudomonas, S.typhi* and *S.aureus* whereas against *B.subtilis* and *E.coli* poor activity was recorded. Due to oximation, compound **5** registered very good activity against *S.aureus, B.subtilis* and *E.coli* whereas against *S.typhi* and *pseudomonas* no improvement in activity was noted.

The addition of another methyl group at C-3 position of compound **5**(compound **6**) showed excellent activity against *S.aureus, B.subtilis, E.coli* and *Pseudomonas* while against *S.typhi* poor activity

was noted. From the MIC values, it is seen that compounds **2** and **6** showed excellent activity against all the tested bacterial strains and which is also comparatively more than that of the reference drug used.

Table 8. HMBC correlation of compound 2.

	Carbon	CH_3	CH_3	C-5	C-3	C-6	C-2	C-4
Proton								
	ppm	21.78	20.84	40.56	50.73	56.71	61.42	212.39
CH_3	0.78	Bonded	β	–	α	–	β	β
CH_3	1.26	β	Bonded	–	α	–	β	β
H-5e	2.57	–	–	Bonded	β	–	α	α
H-5a	3.02	–	–	Bonded	β	α	α	α
H-6	5.08	–	–	β	–	Bonded	β	–
H-2	5.15	β	β	–	–	β	Bonded	–

Table 9. Antibacterial activity of compounds 1-6.

compound	Minimum inhibitory concentration in µg/mL				
	S.aureus	*B.subtilis*	*S.typhi*	*E.coli*	*Pseudomonas*
1	50	100	25	50	200
2	12.5	50	6.25	12.5	12.5
3	50	6.25	50	25	50
4	12.5	200	6.25	100	6.25
5	50	25	100	25	200
6	6.25	12.5	50	50	25
Ciproflaxacin	50	12.5	50	12.5	50

The percentage of antimicrobial potency of the tested compounds compared with reference standard was calculated by adopting the following equation.

Antimicrobial potency (%)

$$= \frac{MIC\left(\frac{\mu g}{mL}\right) of\ reference\ compound}{MIC\left(\frac{\mu g}{mL}\right) of test\ compound} \times 100$$

Antifungal activity

Table 10 exhibits the *in vitro* antifungal studies of compounds **1-6**. Here, amphotericin-B was chosen as standard drug on a list of fungal strains such as *A.niger*, *C.albicans*, *A.flavus*, *Rhizopus sp* and *Mucor*, whose potencies were measured as minimum inhibitory concentration (MIC) (µg/mL).

Table 10. Antifungal activity of compounds **1-6.**

Compound	Minimum inhibitory concentration in µg/mL				
	A.niger	*A.flavus*	*C.albicans*	*Rhizopus sp*	*Mucor*
1	25	25	200	50	25
2	12.5	25	100	25	12.5
3	12.5	12.5	6.25	25	50
4	6.25	50	25	6.25	100
5	50	25	12.5	100	200
6	6.25	12.5	12.5	25	6.25
Amphotericin B	12.5	50	25	50	50

Compound **1** has good activity against *A.flavus*, *Phizopus* and *Mucor* while against *A.niger* and *C.albicans* poor activities were recorded. The further addition of methyl group at C-3 **1**(compound **2**) showed excellent activity against *A.niger*, *A.flavus*, *Rhizopus.sp* and *Mucor* whereas against *C. albicans* better activity was registered. An introduction of ethyl / pentyl group instead of methyl substituent at C-3 position exhibited excellent activities against all the tested fungal strains. Due to oximation, compound **5** showed excellent activity against *A.flavus* and *C.albicans* whereas against *A.niger*, *Rhizopus.sp* and *Mucor* poor activities were noted. Further addition of methyl group at C-3 of compound **5** (compound **6**) exhibited excellent activity against all the tested fungal strains. From the MIC values, it is clearly seen that compounds **3** and **6** showed excellent activity and is comparatively more than that of the reference drug used. The results suggest that the antibacterial and antifungal activities are markedly influenced by substituent in phenyl ring (Figures 2 and 3).

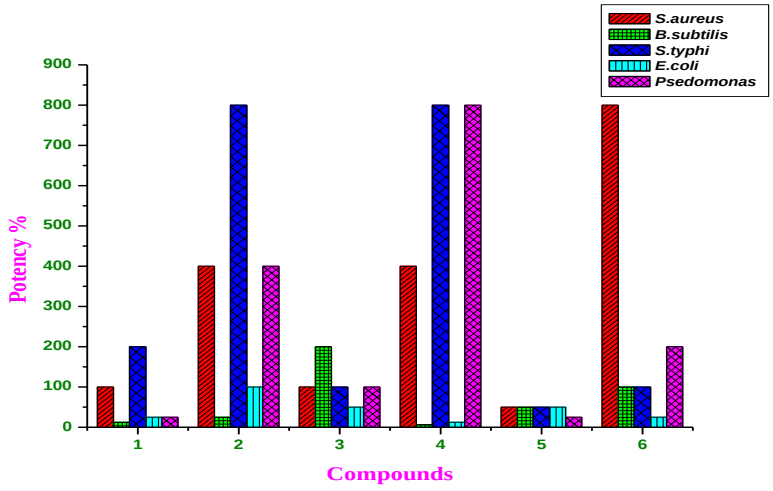

Figure 2. Comparison of antibacterial potency of compounds 1-6 with *Ciprofloxacin*.

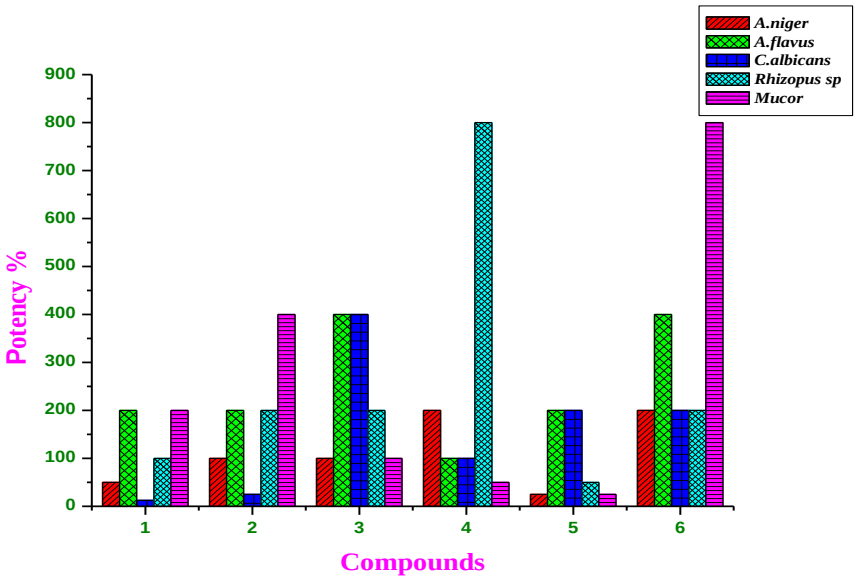

Figure 3. Comparison of antibacterial potency of compounds 1-6 with *Amphotericin B*.

4. CONCLUSIONS

Incorporation of fused phenylgroup at C-2 and C-6 positions of the piperidin-4-one ring makes all the protons deshielded and carbons shielded. This is due to magnetic anisotropic effect. A comparison of chemical shifts of the 3-methylpiperidin-4-one **1** and 3,3-dimethylpiperidin-4-one **2** with those of **5** and **6** reveals that oximation deshields equatorial methylene proton at C-5 but shields axial methylene proton at C-5. A possible explanation based on the steric polarization interaction of the N-O bond with the equatorial *syn* α-(C-H) bond is offered. From ^{13}C chemical shifts of **1-6** it is seen that oximation shields all heterocyclic carbons expect C-2. The magnitude of shielding observed on C-5 in **5** and **6** are greater than that observed on C-3. An explanation based on the lower electronegativtiy of the >C=N group and steric polarization interaction between the N-O bond and the *syn* α-(C-H) equatorial bond is given to rationalize the observations. All the compounds were screened for their preliminary antibacterial and antifungal activities. Compounds **2** and **6** were found to be potent against all the tested microbial organisms, showing MIC 6.25 to 50 µg/mL. Most of the compounds showed good antibacterial and antifungal activities compared to the case where phenyl group is attached to C-2 and C-6 position of piperidine ring.

ACKNOWLEDGMENTS

One of the authors, Dr. G. Rajarajan is thankful to UGC [F. No.42-343/2013 (SR)] for providing funds to this research study. Mr. M. Arockia doss is thankful to UGC for providing fellowship.

REFERENCES

[1] Katritzky, R.; Fan,W.Q. The chemistry of benzotriazole. A novel and versatile synthesis of 1-alkyl-1-aryl-, 1-(alkylamino)-, or 1-1-amido-substituted and of 1,2,6-trisubstituted piperidines from glutaraldehyde and primary amines or monosubstituted hydrazines. *J. Org. Chem*. **1990**, *55*, 3205-3209.

[2] Atta-ur-Rahmanin, S.R.; Angle, J.G. Breitenbucher (Eds), Stereoselective Synthesis, Elesvier, NewYork. **2007**, 453-502.

[3] Pelletier, S.W.; Schneider, M.J. Chemical and biological perspectives, Wiley, New York. **1996** (Eds), 155-355.
[4] Chakkaravarthy, J.; Muthukumaran, G.; Pandiarajan, K. Conformational study of some N-acyl-2r,6c-diphenylpiperidin-4-one oximes using NMR spectra. *J. Mol. Struct*. **2008**, *889*, 297-307.
[5] Parthiban, P.; Balasubramanian, S.; Aridoss, G.; Kabilan, S. Synthesis and NMR spectral studies of some 2,6-diarylpiperidin-4-one O-benzyloximes, *Spectrochim. Acta. A* **2008**, *70*, 1-24.
[6] Ramalingam, C.; Park, Y. T.; Kabilan, S. Synthesis, stereochemistry and antimicrobial evolution of substituted piperidine-4-one oxime ethers. *Eur. J. Med. Chem*. **2006**, *41*, 683-696.
[7] Mokio, I.G.; Soldatenkov, A.T.; Federov, V.O.; Ageev, E.A.; Sergeeva, N.D.; Lin, S.; Stashenku, E.E.; Prostakov, N.S.; Andreeva ,E.L. Antimicrobial activity of aliphatic-aromatic ketones, β-ketols and α-glycols. *Khim Farm Zh*. **1989**, *23*, 421-427.
[8] Parthiban, P.; Aridoss, G.; Rathika, P.; Ramkumar, V.; Kabilan, S. Synthesis, spectral, crystal and antimicrobial studies of biologically potent oxime ethers of nitrogen, oxygen and sulphur heterocycles. *Bioorg. Med. Chem. Lett*. **2009**, *19*, 2981-2985.
[9] Subbagh, H.I. E.L.; Abu-Zaid, S.M.; Mahran, M.A.; Badria, F.A.; Alafaid, A.M. Synthesis and biological evaluation of certain α, β-unsaturated ketones and their corresponding fused pyridines as antiviral and cytotoxic agents. *J. Med. Chem*. **2000**, *43*, 2915-2921.
[10] Watsona, A.A.; Fleet, G.W.J.; Asano, N.; Molyneux, R.J.; Nugh, R.J. Polyhydroxylated alkaloids- natural occurrence andtherapeutic applications. *Phytochemistry*. **2001**, *56*, 265-295.
[11] Aridoss, G.; Amirthaganesan, S.; Kim, M.S.; Kim, J.T.; Jeong, Y.T. Synthesis, spectral and biological evaluation of some thiazolidinones and thiozoles based on t-3-alkyl-r-2,c-6-diarylpiperidin-4-ones. *Eur. J. Med. Chem*. **2009,** *44*, 4199–4210.
[12] Rameshkumar, N.; Veena, A.; Ilavarasan, R.; Adirja, M.; Shanmugapandian, P.; Sridhar, S. K. Synthesis and biological activities of 2,6-diaryl-3-methyl-4-piperidone derivatives. *Biol. Pharm. Bull*. **2003**, *26*, 188-193.
[13] Ganellin C.R.; Spickett, R. G. W. Compounds affecting the central nervous system.1.4-piperidones and related compounds. *J. Med. Chem*. **1965**, *8*, 619-625.
[14] Ramalingam, C.; Balasubramanian, S.; Kabilan, S.; Vasudevan, M. Synthesis study of antibacterial and antifungal activities novel 1-[2-(benzoxazol-2yl)ethoxy]-2,6-diarylpiperidin-4-ones. *Eur. J. Med. Chem*. **2004**, *39*, 527-533.
[15] Balasubramanian, S.; Ramalingam, C.; Aridoss, G.; Kabilan, S. Synthesis and study of antibacterial and antifungal activities of novel 8-methyl-7,9-diaryl-1,2,4,8-tetraazaspiro[4,5] decan-3-thiones. *Eur. J. Med. Chem*. **2005**, *40*, 694-700.
[16] Aridoss, G.; Balasubramanian, S.; Parthiban, P.; Kabilan, S. Synthesis, and *in- vitro* microbiological evaluation of imidazo (4, 5-*b*) pyridinylethoxy piperidones. *Eur. J. Med. Chem*. **2006**, *41*, 268-275.
[17] Aridoss, G.; Parthiban, P.; Ramachandran, R.; Prakash, M.; Kabilan, S.; Jeong, Y.T. Synthesis and spectral characterization of a new class of N-(N-methylpiperazinoacetyl)-2,6-diarylpiperdin-4-one: Antimicrobial, analgesic and antipyretic studies. *Eur. J. Med. Chem*. **2009**, *44*, 577-592.
[18] Aridoss,G.; Balasubramanian, S.; Parthiban,P.; Kabilan, S. Synthesis and NMR spectral studies of N-Chloacetyl-2,6-diarylpiperidin-4-ones. *Spectrachim. Acta A* **2007**, *68*, 1153-1163.
[19] Lakshminarayanan, B.N.; Gnanendra, C.R.; Mahadeva Prasad, T.N.; Sridhar, M.A.; Naik, N.; Chenne, D.; Gowda,; Shashidhara Prasad, J. Structural confonformation of a novel piperidine-4-one derivatives: 1-Acryloyl-3-methyl-2,6-dip-tolyl piperidine 4-one. *J. Chem. Crystallogr*. **2010**, *40*, 686-690.
[20] Vimalraj, V.; Pandiarajan, K.; Ahmad Bhat , I. Synthesis and structural study of 3t-isopropyl-2r,6c-diphenyl-4-oxo piperidinium nitrate and N-benzoyl-3t-isopropyl-2r,6c-diphenyl piperidine-4-one oxime. *J. Mol. Struct*. **2010**, *975*, 137-146.
[21] Jayabharathi, J.; Thangamani, A.; Balamurugan, S.; Thiruvalluvar, A.; Linden, A. *t* -3-Benzyl-*r*-2,*c*-6-bis(4-methoxyphenyl)piperidin-4-one oxime. *Acta. Cryst. E*. **2008**, *64*, 1211.

[22] Venkateshvaramoorthi, R.; Krishnasamy, K. Synthesis, spectral characterization, antimicrobial studies of some 3-substituted piperidine-4-one derivatives. *Int. J. Rec. Sci. Res*. **2013**, *4*, 45-49.
[23] Jayabharathi, J.; Thangamani, A.; Balamurugan, S.; Thiruvalluvar, A.; Linden, A. *t*-3-Benzyl-*r*-2,*c*-6-bis(4-methoxyphenyl)piperidin-4-one. *Acta. Cryst. E*. **2008**, *64*, 1181.
[24] Gopalakrishnan, M.; Thanusu, J.; Kanagarajan, V. J. A Simple and efficient grindstone chemistry solid-state reaction procedure for the synthesis of heterocyclic oximes in dry media. *J. Korean Chem. Soc*. **2008**, *52*, 523-529.
[25] Noller, C.R.; Balliah, V. The preparation of some piperidine derivatives by the Mannich reaction. *J. Am. Chem. Soc*. **1948**, *70*, 3853-3855.
[26] Colapret, J.A.; Diamantidis, G.; Spencer, H.K.; Rudo, F. G.T.C. Synthesis and pharmacological evaluation 4,4-substituted piperidines. *J. Med. Chem*. **1989**, *32*, 968-974.
[27] Rastogi, S.; Rastogi, H. An efficient synthesis of some substituted piperidine-4-one thiosemicarbazone derivatives as potential anti convulsant under microwave irradiation. *Indian J. Chem*. **2010**, *49*, 547-553.
[28] Pandiarajan, K.; Sekar, R.; Anantharaman, R.; Ramalingam, V. Conformational studies of some piperidine-4-ones using PMR spectroscopy. *Indian J. Chem*. **1991**, *30*, 490-493.
[29] Mishah, U.I.; Hasan, M.U.; Mohammad Arab,; Pandiarajan, K.; Sekar, R. Conformational analysis and ^{13}C NMR spectra of some 2,6-diarylpiperidin-4-ones. *Magn. Reson. Chem*. **1985**, *23*, 291-295.
[30] Pandiarajan, K.; Sabapathy Mohan, R.T.; Hasan , M.U. ^{13}C and ^{1}H NMR spectral studies of some piperidin-4-one oximes. *Magn. Reson. Chem*. **1986**, *24*, 312-316.
[31] Savithiri, S.; Rajarajan, G.; Srividhya, V.; Jijesh, J.; Thanikachalam, V.; Jayabharathi , J.; Arockiadoss, M. Synthesis, spectral and structure activity relationship studies of Substituted 3t-pentyl-2r, 6c–diarylpiperidin-4-ones and their corresponding oxime derivatives. *Can. Chem. Trans*. **2014**, *2*, 201-220.

The authors declare no conflict of interest

© 2015 By the Authors; Licensee Borderless Science Publishing, Canada. This is an open access article distributed under the terms and conditions of the Creative Commons Attribution license http://creativecommons.org/licenses/by/3.0