Acta Poloniae Pharmaceutica – Drug Research, Vol. 69 No. 3 pp. 433–438, 2012 ISSN 0001-6837
Polish Pharmaceutical Society

SYNTHESIS AND CHARACTERIZATION OF (Z)-N-(1-[2-{3-[(DIMETHYLAMINO)METHYL)]-2-METHOXYPHENYL}-5-(PYRIDIN-4-YL)-1,3,4-OXADIAZOL-3(2H)-YL]ETHYLIDENE) BENZENAMINE DERIVATIVES AS POTENT ANTIFUNGAL AGENTS

MANAV MALHOTRA[1]*, PANKHURI HANS[1], SAGUN SHARMA[1], AAKASH DEEP[2] and PRIYANKA PHOGAT[3]

[1]Department of Pharmaceutical Chemistry, ISF College of Pharmacy, Ferozepur Road, Moga 142 001, India
[2]Department of Pharmaceutical Sciences, Maharishi Dayanand University, 124001, India
[3]Department of Pharmaceutical Sciences, Hindu College of Pharmacy, Sonepat-131001, India

Abstract: In the present study, a series of (Z)-N-(1-[2-{3-[(dimethylamino)methyl)]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazol-3(2H)-yl]ethylidene)benzenamine derivatives have been synthesized and characterized by IR, ^{1}H NMR and ^{13}C NMR spectra. All the synthesized compounds were evaluated for their antifungal activity and were compared with the standard drug, clotrimazole. The compounds demonstrated excellent to weak antifungal activity. Among the synthesized derivatives, **4f** and **4h** showed significant activity and **4c** exhibited moderate activity against *Candida albicans, Candida tropicalis and Aspergillus niger* as compared with the standard antifungal agent – clotrimazole. The minimum inhibitory concentration of the compounds was in the range of 1.62–25 µg/mL against fungi. Furthermore, the substitution of chloro, nitro and methoxy groups at para position of benzene moiety play an important role in enhancing the antifungal activity of this class of compounds.

Keywords: 1,3,4-oxadiazoles, antifungal activity, hydrazide-hydrazones, Mannich base, lipophilicity

The treatment of infectious diseases still remains an important and challenging problem because of multi-drug resistant microbial pathogens. Resistance to antifungal drugs is the ability of certain microorganisms to withstand attack by potent antifungal agents and this uncontrolled rise in the resistance threatens lives and limits healthcare resources. Life-threatening fungal diseases, which arise as a result of multi-drug resistance, are of great concern in the world. Owing to this increased fungal resistance, novel antifungal agents are needed to combat with the multidrug-resistant fungal diseases (1). Nitrogen containing heterocyclic molecules constitute a great proportion of various chemical entities, which are obtained from many natural products, fine chemicals and biologically active pharmaceuticals. Amongst them 1,3,4-oxadiazole derivatives play an important role in medicinal chemistry, pesticide chemistry, polymer chemistry and act as building blocks in the construction of novel biologically active molecules (2). Because many of 1,3,4-oxadiazoles derivatives display significant biological activity, their synthesis and chemical modification have received particular interest from a long time. Oxadiazoles exhibit wide spectrum of biological activity like antifungal (3, 4), antimicrobial, anti-HIV (5), antitubercular (6), antimalarial (7), analgesic (8), anti-inflammatory (9), anticonvulsant (10), hypoglycemic (11), genotoxicity (12), lipid peroxidase inhibition (13), anticancer (14), tyrosinase inhibition (15) and muscle relaxant properties (16). In addition, derivatives of 1,3,4-oxadiazoles are also used as photosensitizers and liquid crystals (17, 18). Oxadiazole group in various molecules have the ability to reach the specific target by transmembrane diffusion and show potent antifungal action (19). Keeping in mind the importance of oxadiazole nucleus, it was found worth to design and synthesize a series of novel derivatives bearing oxadiazole moiety and screen them for their antifungal activity.

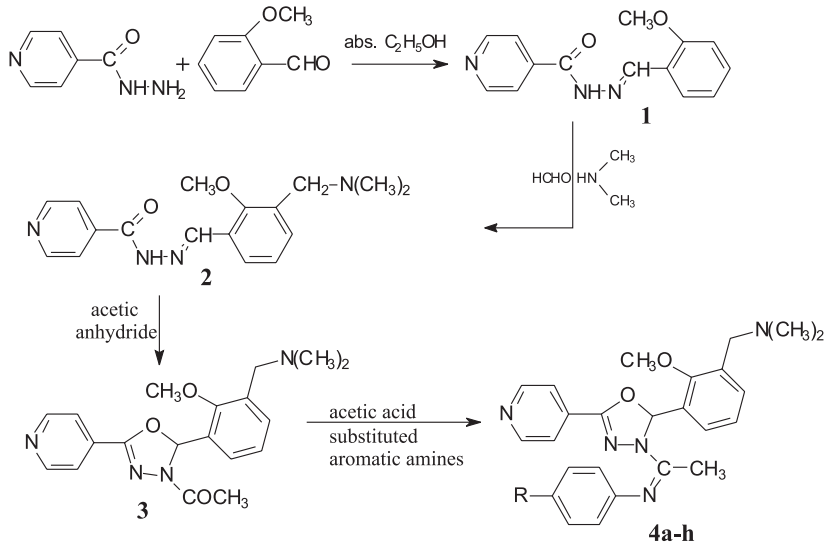
CHEMISTRY

The synthesis of target compounds was carried as outlined in Scheme 1. Compounds (**4a**–**4h**) were

* Corresponding author: e-mail: manavmalhotra99@yahoo.in

Table 1 Physical data of title compounds (**4a-h**).

Comp. No.	R	Molecular formula	Molecular weight	Melting point (°C)	Yield %
4a	H	$C_{25}H_{27}N_5O_2$	429.5	205–207	72
4b	F	$C_{25}H_{26}FN_5O_2$	447.5	212–214	64
4c	Cl	$C_{25}H_{26}ClN_5O_2$	463.9	218–220	69
4d	Br	$C_{25}H_{26}BrN_5O_2$	508.4	215–217	65
4e	I	$C_{25}H_{26}IN_5O_2$	555.4	195–197	58
4f	NO_2	$C_{25}H_{26}N_6O_4$	474.5	217–219	73
4g	CH_3	$C_{26}H_{29}N_5O_2$	443.5	203–205	62
4h	OCH_3	$C_{26}H_{29}N_5O_2$	459.2	210–212	65



Scheme 1. Synthetic pathway for the formation of title compounds

readily prepared with good yield and purity. Equimolar quantities of 2-methoxybenzaldehyde and isonicotinoyl hydrazide were refluxed to form (E)-N'-(2-methoxybenzylidine) isonicotinoyl hydrazide (**1**), which on reaction with formaldehyde and substituted secondary amines forms (E)-N'-{3-[(dimethylamino)methyl]-2-methoxybenzylidene}isonicotinoyl hydrazide (**2**), which on further treatment with excess of acetic anhydride formed 1-[2-{3-[(dimethylamino)methyl]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazole-3(2H)-yl]ethanone (**3**) and at last on reaction with substituted aromatic amines it forms a series of (Z)-N-(1-[2-{3-[(dimethylamino)methyl]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazol-3(2H)-yl]ethylidene)benzenamine derivatives (**4a–h**)**.** All the synthesized compounds (**4a–4h**) were obtained with good yield and the physical and analytical data of synthesized compounds are given in Table 1. The purity of the compounds was checked by TLC, elemental analysis and spectral data. In general, IR spectra of all compounds (**4a–4h**) showed absorption band at 2989–2951, 2865–2839, 1721–1712, 1678–1672, 1574–1563, 1188–1181 and 1085–1055 cm^{-1} regions, conforming the presence of CH, CH_2, C=O, C=N, C=C, C-N and C-O, respectively. In ^1H NMR spectra, the respective derivatives (**4a–h**) were verified on the basis of their chemical shifts, multiplicities, and coupling constants. The spectra of most of the compounds showed a characteristic 4 protons of pyridine signal at δ 8.98–8.25 ppm, characteristic protons of phenyl signal at δ 7.95–6.45 ppm, signal of 1 proton of oxadiazole at δ 5.69–5.46 ppm, 3 protons signal of O-CH_3 at δ 3.88–3.71 ppm, 2 protons signal of Ar-CH_2-N at δ 3.69–2.32 ppm, signal of 6 protons of N-$(CH_3)_2$ at δ 2.69–2.32 ppm

and 3 protons signal of CH_3 at δ 1.05–1.21 ppm. ^{13}C-NMR spectra of compounds (**4a**–**h**) showed characteristic signals which appeared at δ 164.77–164.18 ppm (-N=C-CH_3), δ 161.42-93.25 ppm (phenyl), δ 149.34–124.12 ppm (pyridine), δ 155.25–65.29 ppm (oxadiazole), δ 57.63–55.42 ppm (O-CH_3), δ 55.64–54.18 ppm (Ar-CH_2-N), δ 47.38–46.24 ppm (N-$(CH_3)_2$) and δ 18.46–15.13 ppm (N-C-CH_3).

EXPERIMENTAL

Melting points of the synthesized compounds were determined in open-glass capillaries on Stuart SMP10 melting point apparatus and were uncorrected. The purity of the compounds was checked by thin layer chromatography (TLC). Silica gel (Kiesel gel 0.25 mm, 60 GF_{254}) precoated sheets were obtained from Merck, Darmstadt (Germany) and used for TLC and the spots were visualized using iodine vapors/ultraviolet light as visualizing agent. The IR spectra (δ, cm^{-1}) were obtained with a Perkin-Elmer 1600 FTIR spectrometer in KBr pellets. ^{1}H-NMR spectra (δ, ppm) were recorded in DMSO-d_6 solutions on a Varian-Mercury 300 MHz spectrometer using tetramethylsilane as the internal reference. ^{13}C NMR spectra were recorded in DMSO-d_6 solutions on a Bruker Avance II 400 spectrometer at 400 MHz using tetramethylsilane as the internal reference. Elemental analyses were performed on an ECS 4010 Elemental Combustion System. The necessary chemicals were purchased from Sigma Aldrich Co..

(E)-N'-(2-methoxybenzylidene)isonicotinoyl hydrazide (1)

A mixture of 2-methoxybenzaldehyde (1.36 g, 0.01 mol) and isoniazid (1.37 g, 0.01 mol) in 15 mL of absolute ethanol was refluxed for 7 h. The completion of reaction was confirmed by TLC. The reaction mixture was then poured in ice cold water and the precipitate obtained was filtered and dried in an oven at low temperature. The product was recrystallized from methanol. Yield 68%; m.p. 204–206°C; IR (KBr; cm^{-1}): 3261, 2926, 2865, 2838, 1674, 1652, 1561, 1116, 1064. ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 12.05 (s, 1H, -NH-N=), 8.82 (d, 2H, pyridine, *J* = 4.7 Hz), 8.74 (s, 1H, -N=C-H), 7.88 (d, 2H, pyridine, *J* = 4.7 Hz), 7.82 (d, 1H, benzylidene, *J* = 9.2 Hz), 7.40 (d, 2H, benzylidene, *J* = 8.9), 3.86 (s, 3H, O-CH_3); ^{13}C-NMR (400 MHz, DMSO-d_6, δ, ppm): 163.45, 160.61, 149.81, 143.37, 139.24, 133.58, 131.74, 123.69, 121.31, 117.83, 113.77, 55.44. Analysis: calcd. for $C_{14}H_{13}N_3O_2$: C 65.87, H 5.13, N 16.46%; found: C 65.74, H 5.18, N 16.54%.

(E)-N'-{3-[(dimethylamino)methyl]-2-methoxybenzylidene}isonicotinoyl hydrazide (2)

2-Methoxybenzylideneisonicotinoyl hydrazide (612 mg, 0.0024 mol) along with (0.1 mL, 0.0036 mol) of formaldehyde and (0.0024 mol) of substituted secondary amine (dimethylamine) was placed in 100 mL round bottom flask to which 50 mL of absolute ethanol was added and the pH was adjusted to 4 with hydrochloric acid. The mixture was refluxed for 35 h. The completion of reaction was confirmed by TLC. The reaction mixture was concentrated on water bath and cooled to room temperature and diethyl ether was added. The reaction mixture was kept for 3–5 h in refrigerator, filtered and washed with n-hexane. The product was recrystallized from methanol. Yield 78%; m.p. 222–225°C; IR (KBr; cm^{-1}): 3258, 2952, 2858, 2840, 1668, 1654, 1545, 1121, 1072. ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 11.92 (s, 1H, -NH-N=), 8.74 (d, 2H, pyridine, *J* = 4.2 Hz), 8.44 (s,1H,-N=C-H), 7.85 (d, 2H, pyridine, *J* = 3.9 Hz), 7.54 (d, 2H, benzylidene, *J* = 7.5 Hz), 7.19 (m, 1H, benzylidene), 3.84 (s, 3H, O-CH_3), 3.32 (s, 2H, Ar-CH_2-N), 0.98 (t, 6H, $2CH_3$); ^{13}C-NMR (400 MHz, DMSO-d_6, δ, ppm): 163.59, 160.71, 149.37, 143.45, 139.41, 133.52, 129.82, 122.64, 119.14, 117.38, 113.15, 55.61, 45.57. Analysis: calcd. for $C_{17}H_{20}N_4O_2$: C 65.37, H 6.45, N 17.94%; found: C 65.43, H 6.44, N 17.89%.

1-[2-{3-[(dimethylamino)methyl]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazol-3(2H)yl] ethanone (3)

A mixture of N-{3-[(dimethylamino)methyl]-2-methoxybenzylidene}isonicotinoyl hydrazide (3.57 g, 0.01 mol) with an excess of acetic anhydride was refluxed for 7 h until the completion of the reaction. which was monitored by TLC. The excess of acetic anhydride was distilled off and the residue was poured onto crushed ice. The solid thus obtained was filtered; washed with water and was then recrystallized from aqueous methanol. Yield 75%; m.p. 168–170°C; IR (KBr; cm^{-1}): 2983, 2861, 2842, 1716, 1673, 1565, 1185, 1059. ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 8.94 (d, 2H, pyridine, *J* = 4.6 Hz), 8.34 (d, 2H, pyridine, *J* = 4.2 Hz), 7.35 (d, 2H, phenyl, *J* = 3.7 Hz), 6.69 (m, 1H, phenyl), 5.55 (s, 1H, oxadiazole), 3.69 (s, 3H, O-CH_3), 3.47 (s, 2H, Ar-CH_2-N), 2.18 (s, 6H, N-$(CH_3)_2$), 1.13 (s, 3H, N=C-CH_3); ^{13}C-NMR (400 MHz, DMSO-d_6, δ, ppm): 168.75, 154.88, 154.29, 149.53, 137.91, 126.37, 125.54, 123.91, 119.71, 118.63, 65.75, 55.26, 54.63, 45.12, 28.46. Analysis:

calcd. for $C_{19}H_{22}N_4O_3$: C 64.39, H 6.26, N 15.81%; found: C 64.35, H 6.28, N 15.83%.

General procedure for synthesis of substituted oxadiazoles (4a–h)

A mixture of 0.01 mole of **3** and an equimolar amount of 0.01 mole of appropriate aromatic amine was added to 25 mL of ethanol with catalytic amount of glacial acetic acid (20), and refluxed for 7–9 h to form compounds **4a–h**. The precipitate obtained was filtered, washed with cold methanol and recrystallized from absolute ethanol. Physical properties of synthesized compounds are given in Table 1 and analytical and spectral data were obtained for all the compounds.

(19Z)-N-(1-[2-{3-[(dimethylamino)methyl]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazol-3(2H)-yl]ethylidene)benzenamine (4a)

IR (KBr; cm^{-1}): 2955, 2863, 2841, 1719, 1678, 1571, 1182, 1079. ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 8.98 (d, 2H, pyridine, *J* = 4.8 Hz), 8.19 (d, 2H, pyridine, *J* = 4.2 Hz), 7.35–7.18 (m, 8H, phenyl), 5.69 (s, 1H, oxadiazole), 3.74 (s, 3H, O-CH_3), 3.67 (s, 2H, Ar-CH_2-N), 2.32 (s, 6H, N-$(CH_3)_2$), 1.13 (s, 3H, N=C-CH_3); ^{13}C-NMR (400 MHz, DMSO-d_6, δ, ppm): 164.75, 155.48, 152.17, 149.17, 139.82, 137.15, 129.77, 127.74, 125.18, 124.28, 122.19, 119.75, 117.66, 68.53, 57.63, 54.73, 47.12, 18.46. Analysis: calcd. for $C_{25}H_{27}N_5O_2$: C 69.91, H 6.34, N 16.31%; found: C 69.83, H 6.35, N 16.38%.

(25Z)-N-(1-[2-{3-[(dimethylamino)methyl)]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazol-3(2H)-yl] ethylidene)-4-fluorobenzenamine (4b)

IR (KBr; cm^{-1}): 2967, 2861, 2845, 1717, 1673, 1574, 1188, 1075, 885. ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 8.95 (d, 2H, pyridine, *J* = 4.6 Hz), 8.71 (d, 2H, pyridine, *J* = 4.1 Hz), 7.45 (d, 2H, phenyl, *J* = 3.7 Hz), 7.17 (d, 2H, phenyl, *J* = 3.2 Hz), 6.85–6.67 (m, 3H, phenyl), 5.59 (s, 1H, oxadiazole), 3.88 (s, 3H, O-CH_3), 3.66 (s, 2H, Ar-CH_2-N), 2.45 (s, 6H, N-$(CH_3)_2$), 1.05 (s, 3H, N=C-CH_3); ^{13}C-NMR (400 MHz, DMSO-d_6, δ, ppm): 164.18, 161.42, 155.25, 149.13, 144.54, 138.89, 126.17, 125.58, 124.72, 123.92, 121.11, 120.27, 116.85, 67.54, 56.15, 55.64, 46.28, 15.27. Analysis: calcd. for $C_{25}H_{26}FN_5O_2$: C 67.10, H 5.86, N 4.25%; found: C 66.92, H 5.95, N 4.34%.

(19Z)-N-(1-[2-{3-[(dimethylamino)methyl]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazol-3(2H)-yl]ethylidene)-4-chlorobenzenamine (4c)

IR (KBr; cm^{-1}): 2951, 2859, 2843, 1715, 1673, 1565, 1183, 1074, 1018. ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 8.88 (d, 2H, pyridine, *J* = 4.5 Hz), 8.37 (d, 2H, pyridine, *J* = 4.1 Hz), 7.86 (d, 2H, phenyl, *J* = 3.8 Hz), 7.49 (d, 2H, phenyl, *J* = 3.3 Hz), 6.77-6.59 (m, 3H, phenyl), 5.55 (s, 1H, oxadiazole), 3.79 (s, 3H, O-CH_3), 3.67 (s, 2H, Ar-CH_2-N), 2.39 (s, 6H, N-$(CH_3)_2$), 1.08 (s, 3H, N=C-CH_3); ^{13}C-NMR (400 MHz, DMSO-d_6, δ, ppm): 164.35, 155.44, 154.78, 149.69, 147.18, 138.74, 132.33, 130.29, 127.19, 126.55, 124.37, 122.75, 121.63, 119.68, 66.56, 56.59, 55.27, 47.26, 15.88. Analysis: calcd. for $C_{25}H_{26}ClN_5O_2$: C 64.72, H 5.65, N 15.09%; found: C 64.61, H 5.72, N 15.13.

(19Z)-N-(1-[2-{3-[(dimethylamino)methyl]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazol-3(2H)-yl]ethylidene)-4-bromobenzenamine (4d)

IR (KBr; cm^{-1}): 2972, 2863, 2842, 1721, 1675, 1563, 1185, 1069, 739. ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 8.79 (d, 2H, pyridine, *J* = 4.7

Table 2. *In vitro* antifungal activity of title compounds (**4a–h**).

Compounds	Minimum inhibitory concentration (μg/mL)		
	C. albicans	*C. tropicalis*	*A. niger*
4a	12.5	6.5	25
4b	25	12.5	25
4c	6.5	12.5	12.5
4d	12.5	25	6.25
4e	12.5	25	12.5
4f	3.25	1.62	1.62
4g	25	12.5	25
4h	3.25	3.25	1.62
Clotrimazole (standard drug)	0.20	0.30	0.50

Hz), 8.44 (d, 2H, pyridine, *J* = 4.2 Hz), 7.77 (d, 2H, phenyl, *J* = 3.9 Hz), 7.55 (d, 2H, phenyl, *J* = 3.3 Hz), 6.71–6.48 (m, 3H, phenyl), 5.59 (s, 1H, oxadiazole), 3.71 (s, 3H, O-CH_3), 3.64 (s, 2H, Ar-CH_2-N), 2.34 (s, 6H, N-$(CH_3)_2$), 1.13 (s, 3H, N=C-CH_3); ^{13}C-NMR (400 MHz, DMSO-d_6, δ, ppm): 164.45, 155.39, 154.18, 149.55, 147.64, 137.94, 133.38, 127.95, 125.72, 124.12, 121.72, 119.15, 65.29, 56.74, 55.18, 47.38, 15.34. Analysis: calcd. for $C_{25}H_{26}BrN_5O_2$: C 59.06, H 5.15, N 13.77%; found: C 58.95, H 5.18, N 13.85%.

(25Z)-N-(1-[2-{3-[(dimethylamino)methyl]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazol-3(2H)-yl]ethylidene)-4-iodobenzenamine (4e)

IR (KBr; cm^{-1}): 2975, 2861, 2843, 1715, 1671, 1565, 1181, 1058, 659. ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 8.73 (d, 2H, pyridine, *J* = 4.7 Hz), 8.35 (d, 2H, pyridine, *J* = 4.3 Hz), 7.89 (d, 2H, phenyl, *J* = 3.8 Hz), 7.53 (d, 2H, phenyl, *J* = 3.2 Hz), 6.69–6.57 (m, 3H, phenyl), 5.46 (s, 1H, oxadiazole), 3.72 (s, 3H, O-CH_3), 3.56 (s, 2H, Ar-CH_2-N), 2.42 (s, 6H, N-$(CH_3)_2$), 1.18 (s, 3H, N=C-CH_3); ^{13}C-NMR (400 MHz, DMSO-d_6, δ, ppm): 164.57, 155.48, 154.33, 149.71, 147.29, 137.36, 135.86, 127.75, 126.31, 124.92, 123.21, 120.65, 93.25, 66.17, 56.71, 54.92, 45.91, 15.88. Analysis: calcd. for $C_{25}H_{26}IN_5O_2$: C 54.06, H 4.72, N 12.61%; found: C 54.15, H 4.73, N 12.51%.

(25Z)-N-(1-[2-{3-[(dimethylamino)methyl]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazol-3(2H)-yl]ethylidene)-4-nitrobenzenamine (4f)

IR (KBr; cm^{-1}): 2985, 2858, 2839, 1719, 1673, 1564, 1184, 1055, 589. ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 8.95 (d, 2H, pyridine, *J* = 4.7 Hz), 8.25 (d, 2H, pyridine, *J* = 4.2 Hz), 7.95 (d, 2H, phenyl, *J* = 3.8 Hz), 7.55 (d, 2H, phenyl, *J* = 3.3 Hz), 6.83–6.67 (m, 3H, phenyl), 5.59 (s, 1H, oxadiazole), 3.75 (s, 3H, O-CH_3), 3.62 (s, 2H, Ar-CH_2-N), 2.35 (s, 6H, N-$(CH_3)_2$), 1.13 (s, 3H, N=C-CH_3); ^{13}C-NMR (400 MHz, DMSO-d_6, δ, ppm): 164.72, 155.79, 154.13, 149.75, 146.77, 138.44, 127.92, 125.72, 124.93, 123.12, 122.72, 121.19, 119.88, 66.24, 56.57, 55.19, 47.11, 15.13. Analysis: calcd. for $C_{25}H_{26}N_6O_4$: C 63.28, H 5.52, N 17.71%; found: C 63.11, H 5.65, N 17.75%.

(25Z)-N-(1-[2-{3-[(dimethylamino)methyl]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazol-3(2H)-yl]ethylidene)-4-methylbenzenamine (4g)

IR (KBr; cm^{-1}): 2989, 2865, 2845, 1712, 1675, 1569, 1181, 1083. ^{1}H-NMR (300 MHz, DMSO-d_6, δ ppm): 8.92 (d, 2H, pyridine, *J* = 4.6 Hz), 8.35 (d, 2H, pyridine, *J* = 4.2 Hz), 7.85 (d, 2H, phenyl, *J* = 3.9 Hz), 7.28 (d, 2H, phenyl, *J* = 3.4 Hz), 6.72–6.85 (m, 3H, phenyl), 5.75 (s, 1H, oxadiazole), 3.75 (s, 3H, O-CH_3), 3.67 (s, 2H, Ar-CH_2-N), 2.69 (s, 6H, N-$(CH_3)_2$), 2.35 (s, 3H, CH_3), 1.10 (s, 3H, N=C-CH_3); ^{13}C-NMR (400 MHz, DMSO-d_6, δ, ppm): 164.77, 155.54, 154.28, 149.83, 146.75, 138.56, 137.12, 130.24, 126.55, 125.74, 124.31, 122.18, 119.87, 25.21, 15.81. Analysis: calcd. for $C_{26}H_{29}N_5O_2$: C 70.41, H 6.59, N 15.79%; found: C 70.45, H 6.49, N 15.85%.

(25Z)-N-(1-[2-{3-[(dimethylamino)methyl]-2-methoxyphenyl}-5-(pyridin-4-yl)-1,3,4-oxadiazol-3(2H)-yl]ethylidene)-4-methoxybenzenamine (4h)

IR (KBr; cm^{-1}): 2975, 2863, 2841, 1712, 1672, 1564, 1184, 1085. ^{1}H-NMR (300 MHz, DMSO-d_6, δ ppm): 8.87 (d, 2H, pyridine, *J* = 4.5 Hz), 8.28 (d, 2H, pyridine, *J* = 4.1 Hz), 7.83 (d, 2H, phenyl, *J* = 3.8 Hz), 7.36 (d, 2H, phenyl, *J* = 3.2 Hz), 6.59–6.45 (m, 3H, phenyl), 5.65 (s, 1H, oxadiazole), 3.72 (s, 6H, O-2CH_3), 3.69 (s, 2H, Ar-CH_2-N), 2.49 (s, 6H, N-$(CH_3)_2$), 1.21 (s, 3H, N=C-CH_3); ^{13}C-NMR (400 MHz, DMSO-d_6, δ, ppm): 164.75, 159.23, 155.17, 154.29, 149.27, 140.37, 137.93, 127.24, 126.19, 124.27, 123.17, 120.92, 119.33, 115.55, 55.42, 54.18, 46.24, 15.83. Analysis: calcd. for $C_{26}H_{29}N_5O_3$: C 67.95, H 6.36, N 15.24%; found: C 67.83, H 6.35, N 15.37%.

Antifungal evaluation

Screening of finally synthesized compounds exhibited *in vitro* antifungal activity against fungal strains: *C. albicans*, C. *tropicalis* and *A. niger*, assessed by serial two fold dilution technique. Clotrimazole was used as a standard drug for antifungal activity. All the compounds were dissolved in dimethyl sulfoxide (DMSO) to obtain a concentration of 10 µg/mL. Twofold dilutions of test and standard compounds were prepared in Sabouraud dextrose broth I.P. (21). The stock solution was serially diluted to obtain concentrations of 25–0.78 µg/mL in nutrient broth. The inoculum size was approximately 10^6 CFU/mL. The whole batch was incubated for 7 days at 35°C for *A. niger* and at 25°C for *C. albicans* and *C. tropicalis*. After incubation, the inoculated culture tubes were macroscopically examined for turbidity. The culture tubes showing turbidity (lower concentration) and the culture tubes showing no turbidity (higher concentration) indicated the minimum inhibitory concentration (MIC) for the compounds mentioned in Table 2.

RESULTS AND DISCUSSION

In this study eight compounds of oxadiazole were synthesized and evaluated for their antifungal activity. At first, an acid hydrazone was synthesized which on Mannich reaction form N-{3-[(dimethylamino)methyl]-2-methoxybenzylidene}isonicotinoyl hydrazide. The treatment of Mannich base with acetic anhydride yielded 1-[2-{3-[(dimethylamino)methyl]-2-methoxyphenyl}-5-(pyridine-4-yl)-1,3,4-oxadiazole-3(2H)-yl]ethanone which, on further treatment with acetic acid and substituted aromatic amines, gave the corresponding desired compounds (**4a**–**h**). Their analytical data obtained were in agreement with the calculated values of the proposed structures. The compounds were evaluated for their antifungal properties in comparison with clotrimazole. The results of the synthesized compounds revealed that compounds **4f** and **4h** showed significant activity, while compound **4c** showed moderate activity against *C. albicans, C. tropicalis* and *A. niger* and the rest of compounds were found less active.

CONCLUSION

Fungal infections remain a significant cause of morbidity and mortality despite advances in medicinal chemistry with the emergence of various resistant strains. So, we have tried to explore the combination of Mannich base with oxadiazole moiety in order to explore their synergistic antifungal activity. Thus, compounds **4f** and **4h** showed significant activity and compound **4c** showed moderate activity comparable to clotrimazole, while, the rest of compounds were found to be less active as compared with the standard agent. The present study showed that the synthesized compounds can be used as template for future development through modification and derivatization to obtain more potent and selective agents with activity against resistant strains for the treatment of various fungal infections.

REFERENCES

1. Dolman S.J., Gosselin F., Shea P.D., Davies I.W.: J. Org. Chem. 71, 9548 (2006).
2. Khalil M.A., El-Khawss S.M., Kassem M.G.: Int J. Life Sci. Pharma Res. 48, 344 (1980).
3. Dutta M.M., Goswami B.N., Kataky J.C.S.: J. Heterocycl. Chem. 23, 793 (1986).
4. Xia J., Lu H., Gui Y., Zu X.: J. Agric. Food Chem. 50, 3757 (2002).
5. El-Emam A.A., Al-Deeb O.A., Al-Omar M., Lehmann J.: Bioorg. Med. Chem. 12, 5107 (2004).
6. Kucukguzel S.G., Oruc E.E., Rollas S., Sahin F., Ozbek A.: Eur. J. Med. Chem. 37, 197 (2002).
7. Kagthara P.R., Shah N.S., Doshi R.K., Parekh H.H.: Indian J. Chem. 38, 572 (1999).
8. Akhter M., Husain A., Azad B., Ajmal M.: Eur. J. Med. Chem. 44, 2372 (2009).
9. Unangast P.C., Shrum G.P., Conner D.T., Dyer C.D., Schrier D.J.: J. Med. Chem. 35, 3691 (1992).
10. Khan M.S.Y., Khan R.M., Drabu S.: Indian J. Heterocycl. Chem. 11, 119 (2001).
11. Neal J.B., Rosen H., Russell P.B., Adams A.C., Blumenthal A.: J. Med. Chem. 5, 617 (1962).
12. Maslat A.O., Abussaud M., Tashtoush H., Al-Tilab M.: Pol. J. Pharmacol. 54, 55 (2002).
13. Farghaly A.A., Bekhit A.A., Park J.Y.: Arch. Pharm. (Weinheim) 333, 53 (2000).
14. Kumar D., Sundaree S., Johnson E.O., Shah K.: Bioorg. Med. Chem. Lett. 19, 4492 (2009).
15. Khan M.T.H., Chaudhary M.I., Khan K.M., Rani M., Rahmann A.U.: Bioorg. Med. Chem. 13, 3385 (2005).
16. Vousooghi A.N., Tabatabai S.A., Eezadeh A.K., Shafiee A.: Acta Chim. Slov. 54, 317 (2007).
17. Wang J.F., Jabbour G.E., Eugene A.Mash E.A., Anderson J., Zhang Y., Lee P.A. et al.: Adv. Mater. 11, 1266 (1999).
18. Parra M., Hidalgo P., Carrasco E., Barbera J., Silvino L.: Liq. Cryst. 33, 875 (2006).
19. Testa B., Crivori P., Reist M., Carrupt P.A.: Perspect. Drug Discov. Des. 19, 179 (2000).
20. Chawla R., Arora A., Parameswaran M.K., Sharma P.C., Michael S., Ravi T.K.: Acta Pol. Pharm. Drug Res. 67, 247 (2010).
21. Pharmacopoeia of India, vol. II, p. A-88, Ministry of Health Department, Govt. of India, New Delhi 1996.

Received: 26. 03. 2011