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Research Article

Synthesis and Antiviral Bioassay of New Diphenyl Ether-based Compounds

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A set of diphenyl ether derivatives bearing different heterocycles were synthesized from 4-phenoxybenzohydrazide 1 in good yield. Synthesized compounds were screened against a broad panel of viruses in different cell cultures and some of the synthesized compounds showed promising antiviral properties.

Key words: antiviral, chemical biology, diphenyl ethers, drug discovery, heterocycles

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Viral infections is a devastating and one of the common infectious health problems. Acquired immunodeficiency syndrome (AIDS) caused by the human immunodeficiency virus (HIV) remains a health threat of global significance. Beside HIV, there are several other virus causing various diseases like herpesvirus family that contains eight known human viruses, including herpes simplex virus 1 (HSV-1) and herpes simplex virus 2 (HSV-2). HSV-1 causes stomatitis, encephalitis, and ocular infections, while herpes simplex virus 2 (HSV-2) causes genital lesions. (1) Vesicular stomatitis virus (VSV) causes an economically important disease in cattle, horses, and swine. (2) Varicella-zoster virus (VZV) is the causative agent of chickenpox and shingles. (3) Influenza is another serious public health problem that causes severe illness and death in high-risk populations.[a]

Various antiviral agents are available in the market, but most of them are suffering from some limitations such as limited efficacy, development of resistance, and adverse effects. At present, many researches are going on to search new lead compounds that either target distinct stages of the viral replication cycle in a virus-specific way or have a broader spectrum of activity with minimum adverse effect or without any adverse effect. (4,5) In this context, we have synthesized a set of diphenyl ether attached to various heterocyclic moieties and screened them against a broad panel of viruses.

Biaryl ether derivatives are routinely found as very important structural elements for organic synthesis, (6–8) natural products (9,10), and other biologically active compounds. (11) Etravirine and Rilpivirine are biaryl ether antiviral drugs (Figure 1) that used in the treatment of HIV (12–14) and MK-4965 is under clinical trial for antiviral properties. (15) But these drugs are still having major issue with their side-effects.

Azaheterocycle derivatives are known for their versatile biological properties. The nitrogen atom in the heterocycles may modify the electron distribution inside the scaffold which may lead to a modification of the chemical and physical properties of the compounds (solubility, polar surface area, etc.).

Herein, we designed and synthesized some modified analogs of biaryl ether antiviral agents using 4-phenoxybenzohydrazide **1** as building block. All the synthesized compounds screened for *in vitro* antiviral properties against various viruses.

Experimental Section

All chemicals were purchased from Sigma-Aldrich and Cornell Lab (Cairo, Egypt). ^{1}H and ^{13}C NMR spectra were recorded at 500 and 125 MHz, respectively, on a Varian Inova 500 MHz spectrometer at School of Chemistry, University of Wollongong, Wollongong 2522, Australia. Melting points were determined using a Gallenkamp (Griffin) melting point apparatus. Elemental analyses were carried out at the Micro Analytical Center, Faculty of Science, Cairo University, Cairo, Egypt.

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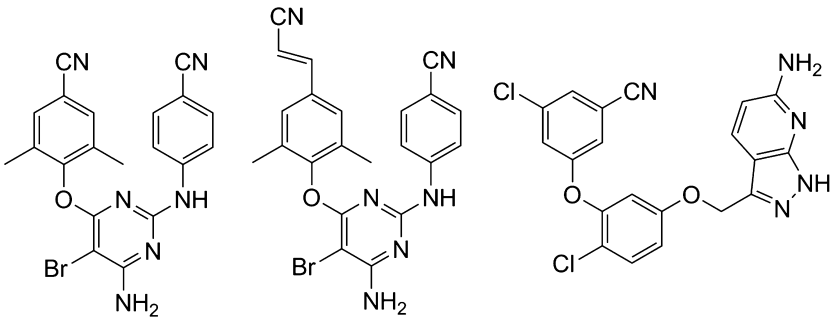


Figure 1: Biaryl ether-derived antiviral agents.

General procedure for preparation of pyrazole/pyrazoline derivatives (2, 3, 4a, b)

To a solution of the acid hydrazide **1** (10 mmol) in 30 mL EtOH containing 2 mL glacial acetic acid, an equimolar amount of 1,3-diketo compounds (acetylacetone, benzoylacetone, or ethyl acetoacetate or ethyl cyanoacetate) was added. The reaction mixture was heated under reflux condition for 12 h. The mixture was cooled, concentrated to a small volume, and added to ice-cold water. The product was filtered, washed with H_2O, and recrystallized from ethanol.

5-Methyl-2-(4-phenoxybenzoyl)-2,4-dihydro-3*H*-pyrazol-3-one (2)

Yield 80%; mp 155–157 °C; ^{1}H NMR (DMSO-d_6) δ 1.98 (s, 3H), 4.17 (s, 2H), 7.08 (d, J = 7.5 Hz, 2H), 7.32 (t, J = 8.0 Hz, 3H), 7.97 (d, J = 8.0 Hz, 2H), 8.02 (d, J = 8.5 Hz, 2H);^{13}C NMR (DMSO-d_6) δ 15.5, 46.2, 117.4, 117.8, 118.4, 121.8, 122.0, 126.9, 127.4, 128.3, 128.9, 152.0, 155.8, 157.3, 164.5, 170.2. Found, %: C, 69.04; H, 4.85; N 9.76. $C_{17}H_{14}N_2O_3$. Calculated, %: C, 69.38; H, 4.79; N, 9.52.

5-Amino-2-(4-phenoxybenzoyl)-2,4-dihydro-3*H*-pyrazol-3-one (3)

Yield 81%; mp 211–213 °C; ^{1}H NMR (DMSO-d_6) δ 3.67 (s, 2H), 6.32 (s, 2H), 7.06 (d, J = 7.5 Hz, 2H), 7.22 (t, J = 8.0 Hz, 3H), 7.45 (d, J = 8.0 Hz, 2H), 7.92 (d, J = 8.5 Hz, 2H);^{13}C NMR (DMSO-d_6) δ 70.2, 116.8, 117.5, 117.9, 118.4, 121.8, 122.0, 152.0, 155.8, 157.3, 160.3, 164.5, 170.0. Found, %: C, 64.94; H, 4.23; N 14.66. $C_{16}H_{13}N_3O_3$. Calculated, %: C, 65.08; H, 4.44; N, 14.23.

(3,5-Dimethyl-1*H*-pyrazol-1-yl)(4-phenoxyphenyl) methanone (4a)

Yield 85%; mp 175–177 °C; ^{1}H NMR (DMSO-d_6) δ 2.17 (s, 3H), 2.49 (s, 3H), 6.27 (s, 1H), 7.02–7.09 (m, 4H), 7.32 (t, J = 8.0 Hz, 1H), 7.45 (t, J = 7.5 Hz, 2H), 7.95 (d, J = 8.5 Hz, 2H); ^{13}C NMR (DMSO-d_6) δ 14.3, 14.6, 111.6, 116.9, 118.4, 121.6, 122.0, 128.3, 129.4, 144.5, 152.0, 155.8, 165.2, 169.4. Found, %: C, 73.61; H, 5.48; N 9.87. $C_{18}H_{16}N_2O_2$. Calculated, %: C, 73.95; H, 5.52; N, 9.58.

(3-Methyl-5-phenyl-1*H*-pyrazol-1-yl)(4-phenoxyphenyl)methanone (4b)

Yield 84%; mp 191–193 °C; ^{1}H NMR (DMSO-d_6) δ 2.36 (s, 3H), 6.37 (s, 1H), 7.08 (d, J = 7.0 Hz, 2H), 7.12–7.24 (m, 2H), 7.32 (t, J = 8.0 Hz, 3H), 7.42 (t, J = 7.5 Hz, 3H), 7.95 (d, J = 8.5 Hz, 2H), 8.04 (d, J = 8.5 Hz, 2H);^{13}C NMR (DMSO-d_6) δ 15.2, 107.4, 117.8, 118.4, 121.6, 122.0, 125.9, 126.4, 127.9, 128.3, 128.9, 129.4, 142.3, 144.5, 152.0, 155.8, 164.5, 165.6. Found, %: C, 77.67; H, 5.43; N 7.97. $C_{23}H_{18}N_2O_2$. Calculated, %: C, 77.95; H, 5.12; N, 7.90.

2-(4-Phenoxybenzoyl)-N-phenylhydrazine-1-carbothioamide (5)

A mixture of phenyl isothiocyanates (1.5 mmol) and acid hydrazide **1** (1 mmol) in anhydrous ethanol was refluxed on a water bath for 3 h. The solid obtained on cooling was filtered, washed with water, and recrystallized from ethanol. Yield 92%; mp 205–207 °C; ^{1}H NMR (DMSO-d_6) δ 7.08 (t, J = 8.5 Hz, 3H), 7.15 (t, J = 7.5 Hz, 2H), 7.22 (t, J = 7.5 Hz, 2H), 7.32 (d, J = 8.0 Hz, 2H), 7.45 (t, J = 7.5 Hz, 3H), 7.98 (d, J = 8.5 Hz, 2H), 9.68 (s, 1H), 9.79 (s, 1H), 10.48 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 117.8, 120.0, 124.9, 125.5, 126.5, 127.7, 128.4, 130.6, 130.8, 139.7, 156.1, 160.4, 165.7. Found, %: C, 66.25; H, 4.34; N 11.76. $C_{20}H_{17}N_3O_2S$. Calculated, %: C, 66.10; H, 4.72; N, 11.56.

5-(4-Phenoxyphenyl)-4-phenyl-4H-1,2,4-triazole-3-thiol (6) (16)

Yield 84%; mp 169–171 °C; ^{1}H NMR (DMSO-d_6) δ 7.02 (d, J = 8.0 Hz, 2H), 7.11 (d, J = 8.0 Hz, 3H), 7.24 (t, J = 6.5 Hz, 2 H), 7.36 (t, J = 8.0 Hz, 2H), 7.46 (t, J = 7.0 Hz, 3H), 7.94 (d, J = 8.5 Hz, 2H), 12.74 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 117.7, 118.3, 120.4, 125.2, 125.7, 127.6, 128.0, 128.4, 128.6, 130.8, 132.1, 133.6, 143.1, 155.6, 161.5, 167.2, 172.3.

General procedure for synthesis of compounds (7a–c)

A mixture of compound **5** (0.01 mol), phenacyl bromide derivatives (0.01 mol), and sodium acetate (0.2 mol) in ethanol (60 mL) was heated under reflux condition for 8–10 h. The mixture was cooled and diluted with water. The precipitate obtained was filtered, dried, and recrystallized using aqueous ethanol.

(*E*)-*N*'-(3,4-Diphenylthiazol-2(3*H*)-ylidene)-4-phenoxybenzohydrazide (7a)

Yield 83%; mp = 169–170 °C; ^{1}H NMR (DMSO-d_6) δ 6.69 (s, 1H), 7.0 (t, J = 7.5 Hz, 2H), 7.12–7.18 (m, 3H), 7.23 (t, J = 7.0 Hz, 2H), 7.36 (t, J = 8.0 Hz, 2H), 7.45 (t, J = 8.5 Hz, 2H), 7.52 (t, J = 7.5 Hz, 2H), 7.62 (d, J = 8.0 Hz, 2H), 7.89 (d, J = 9.0 Hz, 2 H), 7.96 (t, J = 7.5 Hz, 2H), 10.63 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 100.3, 116.9, 118.3, 119.7, 121.7, 121.9, 124.5, 127.6, 128.0, 128.4, 128.6, 129.8, 130.2, 133.4, 133.6, 139.8, 146.3, 155.3, 159.7, 166.4. Found, %: C, 72.41; H, 4.32; N 9.40. $C_{28}H_{21}N_3O_2S$. Calculated, %: C, 72.55; H, 4.57; N, 9.06.

(*E*)-*N*'-(4-(4-Chlorophenyl)-3-phenylthiazol-2(3*H*)-ylidene)-4-phenoxybenzohydrazide (7b)

Yield 85%; mp 169–170 °C; ^{1}H NMR (DMSO-d_6) δ 6.99–7.02 (m, 3H), 7.06–7.14 (m, 6H), 7.24 (t, J = 7.0 Hz, 2H), 7.36 (t, J = 7.5 Hz, 2H), 7.45 (t, J = 8.5 Hz, 2H), 7.61 (d, J = 8.0 Hz, 2H), 7.89 (d, J = 9.0 Hz, 2 H), 10.66 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 104.7, 117.7, 118.4, 119.1, 119.3, 120.5, 122.5, 125.3, 127.6, 128.4, 129.8, 131.0, 139.4, 144.3, 146.2, 156.0, 158.1, 160.4, 160.4, 163.2. Found, %: C, 67.65; H, 4.12; N 8.81. $C_{28}H_{20}ClN_3O_2S$. Calculated, %: C, 67.53; H, 4.05; N, 8.44.

(*E*)-*N*'-(4-(4-Methoxyphenyl)-3-phenylthiazol-2(3*H*)-ylidene)-4-phenoxybenzohydrazide (7c)

Yield 86%; mp 179–181 °C; ^{1}H NMR (DMSO-d_6) δ 3.93 (s, 3H), 6.94 (d, J = 8.5 Hz, 2H), 6.99–7.12 (m, 3H), 7.19 (t, J = 7.5 Hz, 2H), 7.26 (t, J = 7.5 Hz, 2H), 7.35–7.44 (m, 6H), 7.53 (d, J = 8.0 Hz, 2H), 7.92 (d, J = 9.0 Hz, 2 H), 10.67 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 56.4, 106.3, 117.7, 118.4, 119.1, 119.3, 120.5, 122.5, 125.2, 126.6, 128.4, 129.7, 130.9, 139.3, 145.1, 146.3, 155.9, 158.1, 160.4, 160.4, 164.1. Found, %: C, 70.49; H, 4.63; N 8.35. $C_{29}H_{23}N_3O_3S$. Calculated, %: C, 70.57; H, 4.70; N, 8.51.

Synthesis of 5-(4-phenoxyphenyl)-N-phenyl-1,3,4-thiadiazol-2-amine (8)

Concentrated sulfuric acid (1 mL) was added dropwise to compound **7** (0.001 mol) and stirred at room temperature for 30 min. The reaction mixture was poured into ice water and the precipitate was washed with sodium carbonate solution followed by water and recrystallized from ethanol. Yield 92%; mp 201–203 °C; ^{1}H NMR (DMSO-d_6) δ 7.02–7.06 (m, 4H), 7.11–7.20 (m, 4H), 7.25 (t, J = 10.5 Hz, 2H), 7.47 (d, J = 12.0 Hz, 2H), 7.95 (d, J = 15.0 Hz, 2H), 10.41 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 117.9, 118.6, 121.4, 121.9, 124.3, 125.5, 126.1, 128.4, 130.0, 139.6, 153.1, 155.8, 169.4. Found, %: C, 69.60; H, 4.32; N 12.11. $C_{20}H_{15}N_3OS$. Calculated, %: C, 69.54; H, 4.38; N, 12.17.

Synthesis of potassium 2-(4-phenoxybenzoyl) hydrazine-1-carbodithioate (9)

Acid hydrazide **3** was treated with carbon disulfide in ethanolic potassium hydroxide by following the method of Reid and Heindel (17) to afford potassium salt of N'-phenoxybenzoylhydrazine carbodithioic acid. Potassium salt was washed with ether and used in next step without purification.

Synthesis of 4-amino-5-(4-phenoxyphenyl)-4H-1,2,4-triazole-3-thiol (10)

A suspension of potassium dithiocarbazinate 9 (0.1 mole) in water (5 mL) and hydrazine hydrate (15 mL, 0.3 mole) was refluxed for 30 min with occasional shaking. The color of the reaction mixture changed to green with the evolution of hydrogen sulfide gas (lead acetate paper and odor). A homogeneous reaction mixture was obtained during the reaction process. The reaction mixture was cooled to room temperature and diluted with water (100 mL). On acidification with AcOH, the desired triazole was precipitated out. The precipitate was filtered, washed with cold water, and recrystallized from ethanol. Yield 80%; mp 160–162 °C; ^{1}H NMR ($CDCl_3$) δ 4.83 (s, 2H), 7.07–7.11 (m, 4H), 7.20 (t, J = 7.5 Hz, 1H), 7.40 (t, J = 8.0 Hz, 2H), 8.08 (d, J = 8.5 Hz, 2H), 10.59 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 117.6, 118.7, 122.3, 127.3, 127.5, 131.4, 147.6, 158.1, 160.4, 182.5. EI-MS (m/z, 100%): 284 [M^+] (100). Found, %: C, 58.95; H, 4.55; N 20.05. $C_{14}H_{12}N_4OS$. Calculated, %: C, 59.14; H, 4.25; N, 19.70.

General procedure for compound (11a-b)

Method A: To 0.10 mmol KOH in 150 mL of methanol solution were added 0.10 mmol of **10** and then 20 mL of CS_2. After the mixture was heated to reflux for 24 h, the solvent was removed under reduced pressure, the residue was poured into 200 mL of water, and the latter was acidified with conc. HCl to yield a white solid of **11a.** Method B: A mixture of **10** (10 mmol) and thiourea or urea (12 mmol) was fused at 180–190 °C for 3 h and then allowed to cool and washed with water. The solid was filtered and recrystallized from ethanol to give white plates of **11a** or **11b.**

3-(4-Phenoxyphenyl)-[1,2,4]triazolo[3,4-*b*][1,3,4] thiadiazole-6-thiol (11a)

Yield 75%; mp 176–178 °C; ^{1}H NMR (DMSO-d_6) δ 7.13 (d, J = 7.5 Hz, 2H), 7.22 (t, J = 8.0 Hz, 3H), 7.46 (d, J = 7.5 Hz, 2H), 8.22 (d, J = 8.5 Hz, 2H), 10.44 (s, 1H); ^{13}C

NMR (DMSO-d_6) δ 117.4, 117.8, 118.3, 119.4, 119.7, 124.6, 128.9, 130.0, 130.3, 141.2, 154.4, 155.2, 163.8, 181.5. Found, %: C, 55.30; H, 3.13; N 17.45. $C_{15}H_{10}N_4OS_2$. Calculated, %: C, 55.20; H, 3.09; N, 17.17.

3-(4-Phenoxyphenyl)-[1,2,4]triazolo[3,4-*b*][1,3,4] thiadiazol-6-ol (11b)

Yield 73%; mp 213- 215 °C; ^{1}H NMR (DMSO-d_6) δ 7.02 (d, J = 7.5 Hz, 2H), 7.07–7.23 (m, 2H), 7.25 (t, J = 7.5 Hz, 3H), 8.13 (d, J = 8.5 Hz, 2H), 10.42 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 117.9, 118.4, 119.9, 123.4, 128.9, 129.4, 130.4, 149.9, 155.1, 157.8, 163.8. EI-MS (m/z, 100%): 310 [M$^+$] (100). Found, %: C, 58.01; H, 3.21; N 18.26. $C_{15}H_{10}N_4O_2S$. Calculated, %: C, 58.06; H, 3.25; N, 18.05.

General procedure for compound (12a-b)

A mixture of **10** (0.005 mol) and a cyano compound RCH_2CN (0.005 mol) in glacial AcOH (25 mL) and a catalytic amount of conc. H_2SO_4 was refluxed for 3 h and then, after cooling, was diluted with H_2O (20 mL) and neutralized with NH_3 solution. The crude product thus obtained was collected by filtration, washed with H_2O, and crystallized (EtOH) to give **12a,b** as colorless crystals in 62–80% yield.

6-Methyl-3-(4-phenoxyphenyl)-[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazole (12a)

Yield 80%; mp 180–182 °C; ^{1}H NMR (DMSO-d_6) δ 2.98 (s, 3H), 7.02 (d, J = 7.5 Hz, 2H), 7.09–7.24 (m, 2H), 7.25 (t, J = 7.5 Hz, 3H), 8.33 (d, J = 8.5 Hz, 2H); ^{13}C NMR (DMSO-d_6) δ 21.3, 118.2, 118.6, 120.4, 123.4, 128.7, 129.6, 131.4, 149.9, 155.3, 157.8, 166.8. Found, %: C, 61.99; H, 3.54; N 18.25. $C_{16}H_{12}N_4OS$. Calculated, %: C, 62.32; H, 3.92; N, 18.17.

2-{3-(4-Phenoxyphenyl)-[1,2,4]triazolo[3,4-*b*][1,3,4] thiadiazol-6-yl}acetamide (12b)

Yield 62%; mp 210–212 °C; ^{1}H NMR (DMSO-d_6) δ 3.96 (s, 2H), 6.94 (d, J = 7.5 Hz, 2H), 7.09–7.24 (m, 2H), 7.25 (t, J = 7.5 Hz, 3H), 7.91 (s, 2H), 8.04 (d, J = 8.5 Hz, 2H); ^{13}C NMR (DMSO-d_6) δ 37.2, 117.8, 118.3, 121.4, 123.5, 128.7, 129.7, 131.6, 149.9, 155.3, 157.8, 166.8, 167.3, 169.9. Found, %: C, 58.37; H, 3.62; N 20.08. $C_{17}H_{13}N_5O_2S$. Calculated, %: C, 58.11; H, 3.73; N, 19.93.

Synthesis of 5-(4-phenoxyphenyl)-1,3,4-thiadiazole-2-thiol (13)

Potassium salt **11** (0.05 mol) was added portion-wise to 98% H_2SO_4 (15 mL) and the resulting clear solution was stirred at room temperature for 4 h. The mixture was cautiously added to crushed ice, stirred for 20 h, and refrigerated for 2 h, and the precipitates were filtered, washed with water, dried, and finally recrystallized from ethanol to furnish the desired compound. Yield 90%; mp 223–225 °C; ^{1}H NMR (DMSO-d_6) δ 7.01–7.21 (m, 3H), 7.24 (d, J = 8.5 Hz, 2H), 7.44 (t, J = 8.0 Hz, 2H), 7.85 (d, J = 8.0 Hz, 1H), 7.92 (d, J = 9.0 Hz, 1H), 11.89 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 117.7, 118.4, 122.1, 123.3, 123.5, 130.0, 133.4, 135.6, 158.1, 160.4, 165.1, 182.5. Found, %: C, 58.52; H, 3.33; N 9.67. $C_{14}H_{10}N_2OS_2$. Calculated, %: C, 58.72; H, 3.52; N, 9.78.

Synthesis of 2-(4-phenoxyphenyl)-1,3,4-oxadiazole (14)

Hydrazide **1** (1 mmol) and triethyl orthoformate (2 mL) were refluxed for 3 h. The excess ester was evaporated; the residue was cooled and triturated with hexane. The solid was filtered off and recrystallized from 1:1 ethanol–water. White solid, yield 73%; mp 140–144 °C; ^{1}H NMR (DMSO-d_6) δ 7.19–7.23 (m, 4H), 7.25 (t, J = 7.5 Hz, 1H), 7.45 (t, J = 7.5 Hz, 2H), 7.08 (d, J = 8.2 Hz, 2 H), 9.01 (s, 1H);^{13}C NMR (DMSO-d_6) δ 117.8, 118.3, 119.9, 124.7, 128.9, 130.3, 154.2, 155.1, 160.2, 163.3. Found, %: C, 70.50; H, 4.48; N 11.37. $C_{14}H_{10}N_2O_2$. Calculated, %: C, 70.58; H, 4.23; N, 11.76.

General procedure for preparation of Hydrazones (15a–c)

A solution of hydrazide **3** (10 mmol) in ethanol (40 ml) was refluxed with various aldehydes (10 mmol) for 3–6 h. The excess of solvent was removed under reduced pressure. After cooling to room temperature, a white solid appeared. This crude product was filtered, washed with diethyl ether, dried, and recrystallized from ethanol.

(*E*)-*N'*-(Furan-2-ylmethylene)-4-phenoxybenzohydrazide (15a)

White solid, yield 89%; mp 192–194 °C; ^{1}H NMR (DMSO-d_6): δ 6.63 (t, J = 5.4 Hz, 1H), 6.92 (d, J = 7.5 Hz, 2H), 7.11 (t, J = 8.0 Hz, 4H), 7.22 (t, J = 7.5 Hz, 1H), 7.45 (t, J = 7.0 Hz, 2H), 7.94 (d, J = 8.0 Hz, 2H), 8.35 (s, 1H), 11.76 (s, 1H);^{13}C NMR (DMSO-d_6) δ 112.3, 117.5, 119.6, 124.4, 127.9, 129.8, 130.3, 137.3, 145.1, 149.5, 155.5, 159.9, 162.3. Found, %: C, 70.37; H, 4.32; N 9.50. $C_{18}H_{14}N_2O_3$. Calculated, %: C, 70.58; H, 4.61; N, 9.15.

(*E*)-4-Phenoxy-*N'*-(thiophen-2-ylmethylene) benzohydrazide (15b)

White solid, yield 92%; mp 182–183 °C; ^{1}H NMR (DMSO-d_6): δ 7.08–7.14 (m, 5H), 7.22 (t, J = 7.0 Hz, 1H), 7.44 (d, J = 7.5 Hz, 3H), 7.65 (d, J = 7.0 Hz, 1H), 7.94 (d, J = 8.5 Hz, 2H), 8.68 (s, 1H), 11.78 (s, 1H); ^{13}C NMR (DMSO-d_6) δ: 117.5, 119.6, 124.4, 127.8, 127.9, 128.8, 129.8, 130.2, 130.3, 133.8, 139.2, 142.7, 159.9, 162.3. Found, %: C, 67.36; H, 3.39; N 9.04. $C_{18}H_{14}N_2O_2S$. Calculated, %: C, 67.06; H, 4.38; N, 8.69.

(*E*)-4-Phenoxy-*N*'-(pyridin-4-ylmethylene) benzohydrazide (15c)

White solid, yield 90%; mp 149–151 °C; ^{1}H NMR (DMSO-d_6) δ 7.08 (d, J = 7.4 Hz, 2H), 7.17–7.21 (m, 3H), 7.44 (t, J = 9.0 Hz, 2H), 7.80 (d, J = 11 Hz, 2H), 7.98 (d, J = 14 Hz, 2H), 8.65 (s, 1H), 8.64 (d, J = 10.3 Hz, 2H), 11.84 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 117.6, 118.3, 118.3, 120.5, 121.6, 125.7, 126.8, 127.5, 142.5, 145.6, 148.8, 157.4, 160.92, 164.62. Found, %: C, 71.46; H, 4.54; N 13.55. $C_{19}H_{15}N_3O_2$. Calculated, %: C, 71.91; H, 4.76; N, 13.24.

Synthesis of 1,3,4-oxadiazole derivatives (16a–d)

An equimolar mixture of hydrazide **1** (0.0054 mol) with various carboxylic acids (0.0054 mol) was refluxed with phosphorus oxychloride (5 mL) for 2–3 h on water bath. Reaction mixture was cooled to room temperature and poured into ice water. The precipitate obtained was filtered off, washed with water, and further purified by recrystallization with ethanol to give 1,3,4-oxadiazoles **16a-d.**

2-(Furan-2-yl)-5-(4-phenoxyphenyl)-1,3,4-oxadiazole (16a)

White solid, yield 77%; mp 129–132 °C; ^{1}H NMR (DMSO-d_6) δ 6.63 (t, J = 5.4 Hz, 1 H), 7.09 (d, J = 7.5 Hz, 2H), 7.18 (d, J = 7.5 Hz, 1H), 7.23–7.25 (m, 3H), 7.39 (t, J = 8.2 Hz, 2H), 7.98 (d, J = 8.0 Hz, 1H), 8.83 (d, J = 8.0 Hz, 2H); ^{13}C NMR (DMSO-d_6) δ 112.3, 114.4, 115.1, 118.3, 120.5, 127.3, 129.4, 139.2, 145.9, 156.8, 157.4, 162.92, 165.62. Found, %: C, 71.36; H, 4.39; N 9.55. $C_{18}H_{12}N_2O_3$. Calculated, %: C, 71.05; H, 4.67; N, 9.21.

2,5-Bis(4-phenoxyphenyl)-1,3,4-oxadiazole (16b)

White solid, yield 82%; mp 161–163 °C; ^{1}H NMR (DMSO-d_6) δ 7.12 (d, J = 8.7 Hz, 4H), 7.25–7.28 (m, 6H), 7.32 (t, J = 9.0 Hz, 4H), 8.23 (d, J = 8.0 Hz, 4H); ^{13}C NMR (DMSO-d_6) δ 113.6, 117.5, 120.5, 125.6, 128.6, 152.07, 154.0, 164.62 (18).

2-(4-Phenoxyphenyl)-5-(thiophen-2-yl)-1,3,4-oxadiazole (16c)

White solid, yield 75%; mp 139–141 °C; ^{1}H NMR (DMSO-d_6) δ 6.94 (d, J = 8 Hz, 2 H), 7.11 (d, J = 7.0 Hz, 2H), 7.21–7.24 (m, 4H), 7.40 (t, J = 8.2 Hz, 2H), 7.70 (d, J = 6.0 Hz, 1H), 7.95 (d, J = 8.0 Hz, 1H); ^{3}C NMR (DMSO-d_6) δ 114.4, 115.1, 118.3, 120.5, 127.3, 127.8, 128.2, 129.4, 132.3, 133.2, 156.8, 157.4, 162.92, 165.62. Found, %: C, 67.35; H, 3.36; N 9.02. $C_{18}H_{12}N_2O_2S$. Calculated, %: C, 67.48; H, 3.78; N, 8.74.

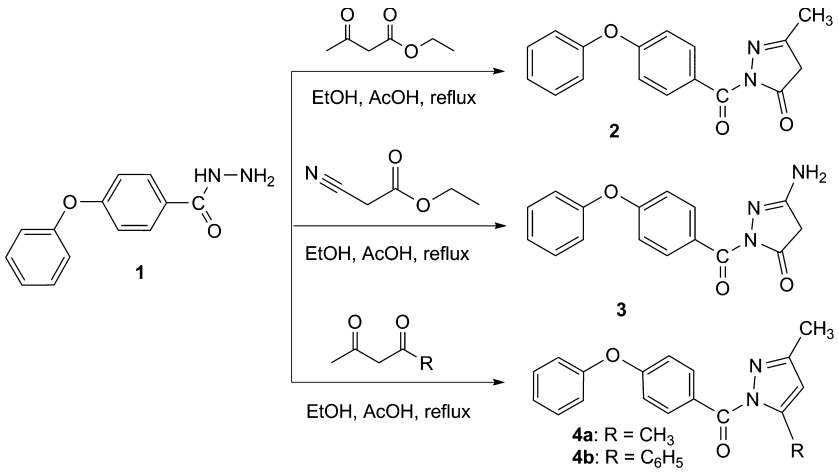
2-(4-Phenoxyphenyl)-5-(pyridin-4-yl)-1,3,4-oxadiazole (16d)

White solid, yield 70%; mp 143–144 °C; ^{1}H NMR (DMSO-d_6) δ 7.03 (d, J = 7.5 Hz, 2H), 7.06 (d, J = 8.2 Hz, 2H), 7.19–7.22 (m, 3H), 7.39 (t, J = 9.0 Hz, 2H), 7.98 (d, J = 7.5 Hz, 2H), 8.83 (d, J = 8.0 Hz, 2H); ^{13}C NMR (DMSO-d_6) δ 114.9, 115.1, 118.3, 120.5, 121.6, 123.5, 127.3, 129.4, 131.2, 132.45, 151.07, 162.92, 165.62. Found, %: C, 72.66; H, 4.39; N 13.65. $C_{19}H_{13}N_3O_2$. Calculated, %: C, 72.37; H, 4.16; N, 13.33.

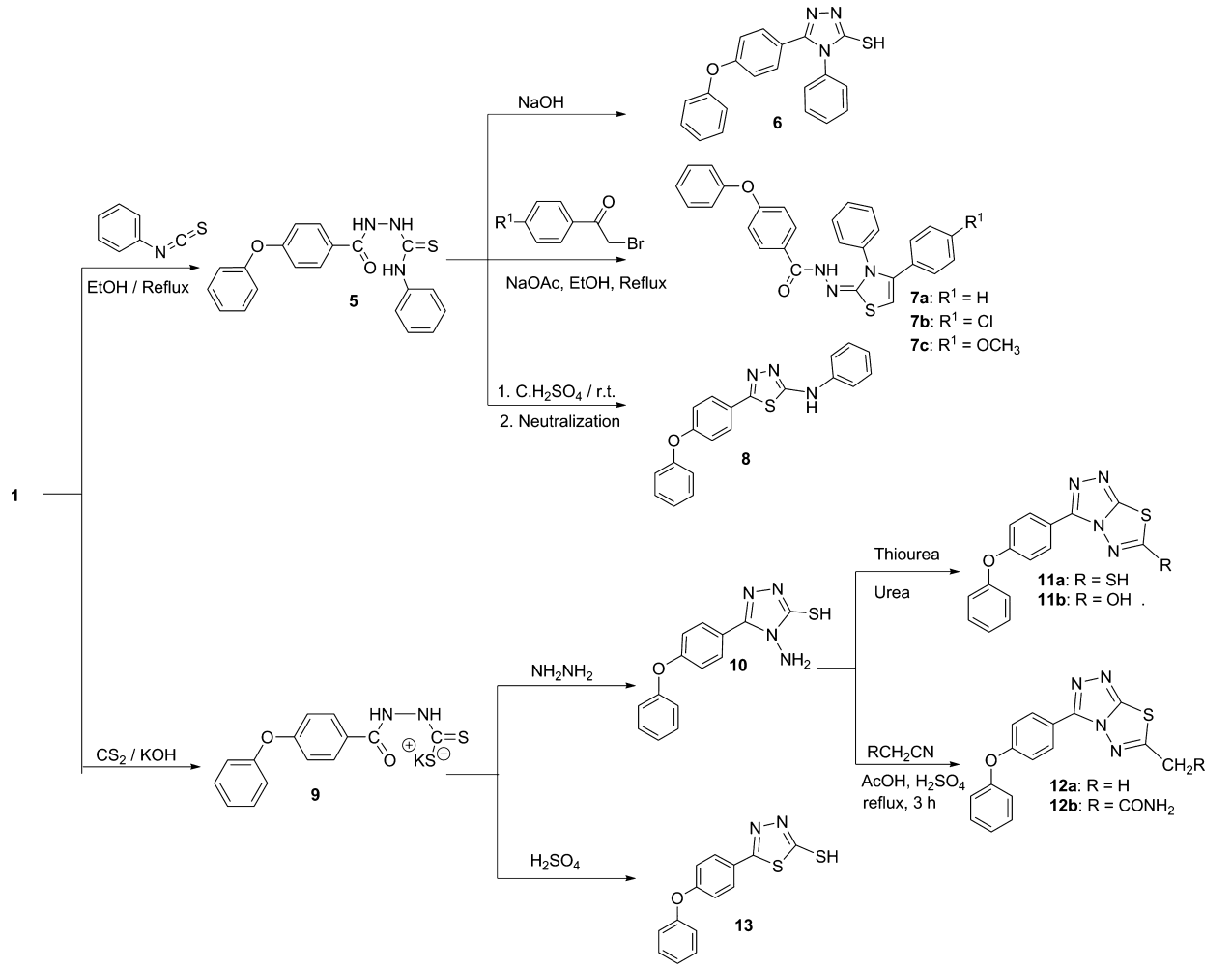
Results and Discussion

Compound **1,** which was synthesized by following the reported method (16), was used as key intermediate because it is a building block for various biological active compound and is the basic moiety of some antiviral agents. (19–22) Compound **3** screened against various viruses and it showed antiviral activity at micromolar concentration.

Cyclocondensation of compound **1** with an appropriate β-dicarbonyl compounds in ethanol using catalytic amount of acetic acid gives the corresponding pyrazole derivatives **2** and **4a,b,** respectively, while cyclocondensation of compound **1** with ethyl cyanoacetate yields compound **3** (Scheme 1).



Scheme 1: Synthesis of compounds **2**, **3,** and **4a,b**.

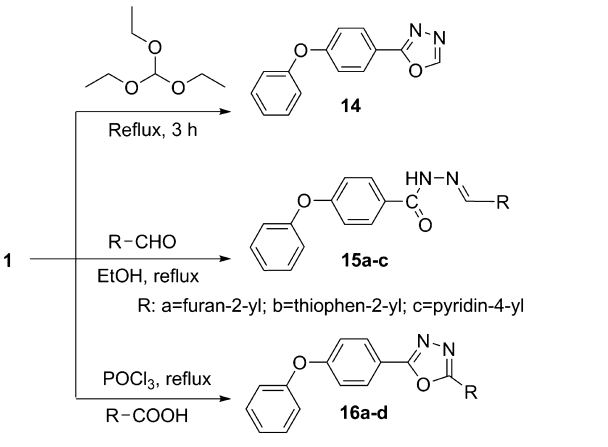


Scheme 2: Synthesis of compounds **7a-c**, **8**, **10**, **11a,b**, **12a,b,** and **13**.

Thiosemicarbazide **5** was afforded by refluxing acid hydrazide **1** with phenyl isothiocyanate in ethanol. Cyclization of thiosemicarbazide **5** using 2N NaOH in ethanol yields the triazole derivative **6**. On the other hand, heating thiosemicarbazide **5** with different phenacyl bromide derivatives and sodium acetate in ethanol yields the corresponding thiazole derivatives **7a-c**, while stirring compound **5** with sulfuric acid gives thiadiazole ring **8** (Scheme 2).

Furthermore, compound **1** stirred in ethanolic potassium hydroxide followed by the addition of carbon disulfide gave potassium salt of N′-phenoxybenzoyl-hydrazine carbodithioic acid **9** which was cyclized to 4-amino-5-(4-phenoxyphenyl)-4H-1,2,4-triazole-3-thiol **10** by refluxing with hydrazine hydrate. Fused heterocyclic derivatives **11a,b** and **12a,b** were prepared by reacting compound **10** with thiourea, urea, acetonitrile, or cyanoacetamide, respectively (Scheme 3). 5-(4-Phenoxyphenyl)-1,3,4-thiadiazole-2-thiol **13** was prepared by stirring **9** with ice-cooled sulfuric acid (Scheme 2).

Finally, we synthesized diphenyl ether bearing oxadiazole, thiophene, furan, and pyridine heterocyclic derivatives.



Scheme 3: Synthesis of compounds **14**, **15a-c,** and **16a-d**.

Cyclization of compound **1** with triethyl orthoformate gives 2-(4-Phenoxyphenyl)-1,3,4-oxadiazole **14**. In addition, condensation of compound **1** with different

Table 1: Antiviral activities of the synthesized compounds

	EC_{50}[a] (μM)											
	Herpes simplex					Influenza			Cytomegalovirus		Varicella-zoster virus	
Compounds	virus 1 (KOS)	virus 2 (G)	Vaccinia virus	Vesicular stomatitis virus	Feline Herpes Virus	A H1N1	A H3N2	B	AD-169 strain	Davis strain	TK^+ OKA	TK^- 07-1
1	9	9	9	>100	13.6	>20	>20	>20	8.94	10.94	10.7	12.3
3	>20	>20	>20	>20	>100	>20	>20	>20	>20	>20	3.35	>20
4a	>100	>100	>100	>100	>100	>100	>100	>100	63.1	100	53.1	45.8
8	>20	>20	>20	>20	>100	5.8	>20	>20	>20	>20	>20	>20
10	73	100	>100	>100	>100	>20	>20	>20	>20	>20	62.7	>20
11a	>20	>20	>20	>20	>100	0.8	0.8	0.8	>20	>20	>20	>20
11b	>20	>20	>20	>20	>100	>20	>20	>20	20	>20	>20	>20
12b	>20	>20	>20	>20	>20	>20	>20	>20	8.94	8.18	>20	>20
13	9	10	9	9	>20	0.8	0.8	0.8	>20	>20	>20	>20
15b	>20	>20	>20	>20	>100	>20	>20	>20	>20	>20	0.8	0.8
16b	>100	>100	>100	45	>100	>100	>100	>100	>20	>20	>20	>20
Cidofovir	2	1.2	17	>250	–	–	–	–	0.22	0.16	–	–
Ganciclovir	0.032	0.032	>100	>100	13.7	–	–	–	1.52	1.20	–	–
Zanamivir	–	–	–	–	–	0.7	10.4	3.2	–	–	–	–
Acyclovir	–	–	–	–	–	–	–	–	–	–	1.34	29.3

[a]Required to reduce virus-induced cytopathogenicity by 50%.

aromatic aldehydes in ethanol yields new hydrazone derivatives **15a-c,** while refluxing of compound **1** with substituted aromatic acids in phosphorous oxychloride leads to the substituted 1,3,4-oxadiazole derivatives **16a-d** (Scheme 3).

Antiviral Bioassay

Screening of this focus library over 17 viruses [herpes simplex virus type 1 (KOS), herpes simplex virus type 2 (G), vaccinia virus, vesicular stomatitis virus, thymidine kinase-deficient herpes simplex virus type 1 (TK- KOS ACVr), para-influenza 3 virus, reovirus-1, Sindbis virus, Coxsackie virus B4, Punta Toro virus, feline corona virus (FIPV), feline herpesvirus, influenza A H1N1 subtype, influenza A H3N2 subtype, influenza B, cytomegalovirus (AD-169 strain and Davis strain), and varicella-zoster virus [TK^+ VZV strain and TK^- VZV strain] leads to demonstrate the effect of various heterocyclic moieties on inhibition of virus-induced cytopathogenicity (19–22) (Table 1, Tables S2–S7).

Compound **11a** showed selectively significant activity against A H1N1, A H3N2, and B influenza viruses. However, compound **13** showed promising broad-spectrum activity against various viruses (herpes simplex, vaccinia virus, vesicular stomatitis virus, and influenza virus). Compound **15b** was selectively active against varicella-zoster virus. Compounds **3** and **8** were found moderate activity against TK^+ and A H1N1, respectively. Rest of the other compounds were possessed mild to negligible antiviral activity. The cytotoxicity of the compounds was evaluated in parallel with their antiviral activity in uninfected cell cultures and is expressed as the minimum cytotoxic concentration (MCC) that causes a microscopically detectable alteration of normal cell morphology (Table S1).

Conclusion

A series of novel diphenyl ether substituted with different heterocyclic rings were synthesized in good yield. The effects of various heterocyclic moieties were studied for antiviral properties. Among all the synthesized compounds **11a**, **13,** and **15b** were more significantly active. The observed bioassay and cytotoxicity data will serve as efficient guide for the future design of potential antiviral agents.

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Note

[a]Influenza (Seasonal) Fact Sheet, World Health Organization, Geneva, 2009. http://www.who.int/mediacentre/factsheets/fs211/en/ (accessed 01.11.12).