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# Synthesis and *In Vitro* Evaluation of Some Novel Benzofuran Derivatives as Potential Anti-HIV-1, Anticancer, and Antimicrobial Agents*

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A novel series of 1-(1-benzofuran-2-yl-ethylidene)-4-substituted thiosemicarbazides (**2a-d**) along with some derived ring systems: substituted-2,3-dihydro-thiazoles (**3a-c, 4a-f**) and thiazolidin-4-ones (**5a-d** and **6a-d**), were synthesized. In addition, cyanoacetic acid-(1-benzofuran-2-yl-ethylidene) hydrazide (**7**) was used to prepare another new series of compounds consisting of substituted pyridin-2(1*H*)-ones (**8a-c**); 2-thioxo-2,3-dihydro-thiazoles (**9a-d**) and 2-thioxo-2,3-dihydro-6*H*-thiazolo[4,5-*d*]pyrimidin-7-ones (**10a-c**, **11a-c**). The absolute configuration of compound **5c** was determined by X-ray crystallography. The compounds prepared were evaluated for their *in vitro* anti-HIV, anticancer, antibacterial, and antifungal activities. Among the tested compounds, compounds **5c** and **9a** produced a significant reduction 비교 the viral cytopathic effect (93.19% and 59.55%) at concentrations >$2.0\times10^{-4}$ M and $2.5\times10^{-5}$ M respectively. Compound **9a** was confirmed to have moderate anti-HIV activity. Compounds **2a**, **2d**, and **5c** showed mild antifungal activity. However, none of the tested compounds showed any significant anticancer activity.

**Key words:** 2-Acetylbenzofuran, Thiosemicarbazides, Caynoacetic acid hydrazide, Dihydro-thiazole, Pyridin-2(1*H*)-ones, Thiazolo[4,5-*d*]pyrimidines, Anti-HIV-1, Anticancer, Antimicrobial activity

## INTRODUCTION

Cancer and AIDS have long been recognized as the most common cause of death. Accordingly, many diverse strategies have been employed to develop new therapies or to improve existing treatments.

A previous literature survey described a variety of 2-substituted benzofuran derivatives that received a great deal of attention for their anti-HIV-1 (Varvaresou *et al.*, 2001; Vacca *et al.*, 1999; Romero *et al.*,1996), anticancer (James *et al.*, 2002; Vinih *et al.*, 2001; Wang *et al.*, 2000; Whomsly, 1993), and antimicrobial (Murat *et al.*, 2005; Wahab Khan *et al.*, 2003; Kawasaki *et al.*, 2002) activities. For example, 1-[(benzofuran-2-yl)phenylmethyl]imidazoles (I) had a potent, reversible, non-selective aromatase-inhibitory effect (Whomsley *et al.*, 1993), 2-{4-[(benzofuran-2-yl)carbonyl]piprazin-1-yl}-3-propylpyridine (II) exhibited good anti-HIV activity (Romero *et al.*, 1996) and 2-(benzimidazol-2-yl-carbonyl)benzofuran derivative (III) (Fig. 1) had a strong inhibitory activity against *Candida albicans N*-myristoyltransferase (Kawasaki *et al.*, 2003).

Many reviews have examined a wide variety of substituted thiazoles and antimetabolites thiazolo[4,5-d]pyrimidines for either their anticancer (Holla *et al.*, 2004; Fahmy *et al.*, 2002, 2003), antiviral (Kini *et al.*, 1991; Negahara *et al.*, 1990) or antimicrobial (Abouzid *et al.*, 2004; Bekhit *et al.*, 2003) activities. In addition, some 2-pyridinone derivatives have been reported to be potent and selective non-nucleoside inhibitors of HIV-1 Reverse transcriptase (Hoffman *et al.*, 1993).

As a part of an ongoing study of biologically active 2-substituted benzofurans, this investigation was directed to synthesize some novel 2-substituted benzofuran derivatives that are structurally related to the previously mentioned

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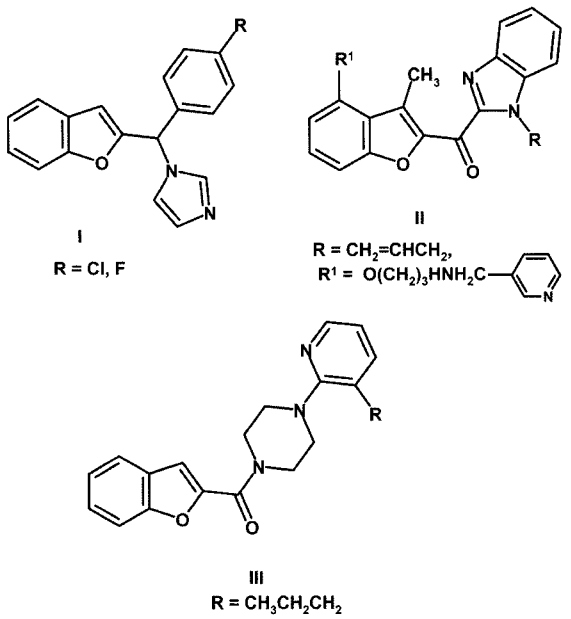
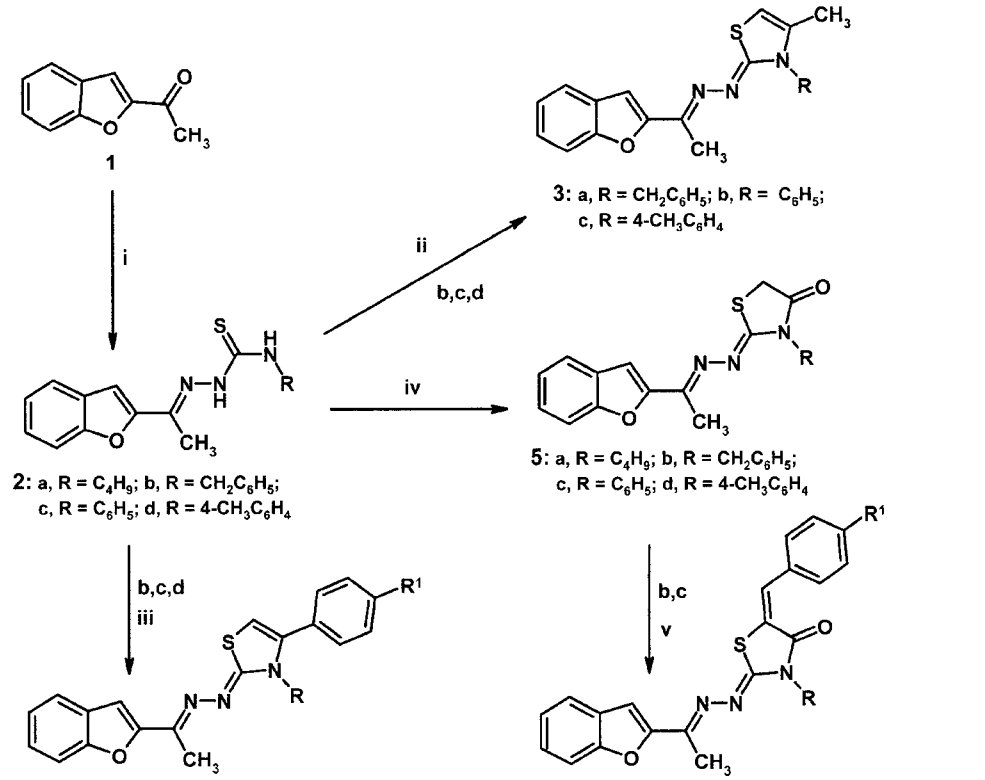



**Fig. 1.** Structures of known 2-substituted benzofuran derivatives with anti-HIV-1, antitumor or antimicrobial activity

biologically active compounds (Fig. 1), and evaluate their *in vitro* anti-HIV-1, anticancer, and antimicrobial activities.

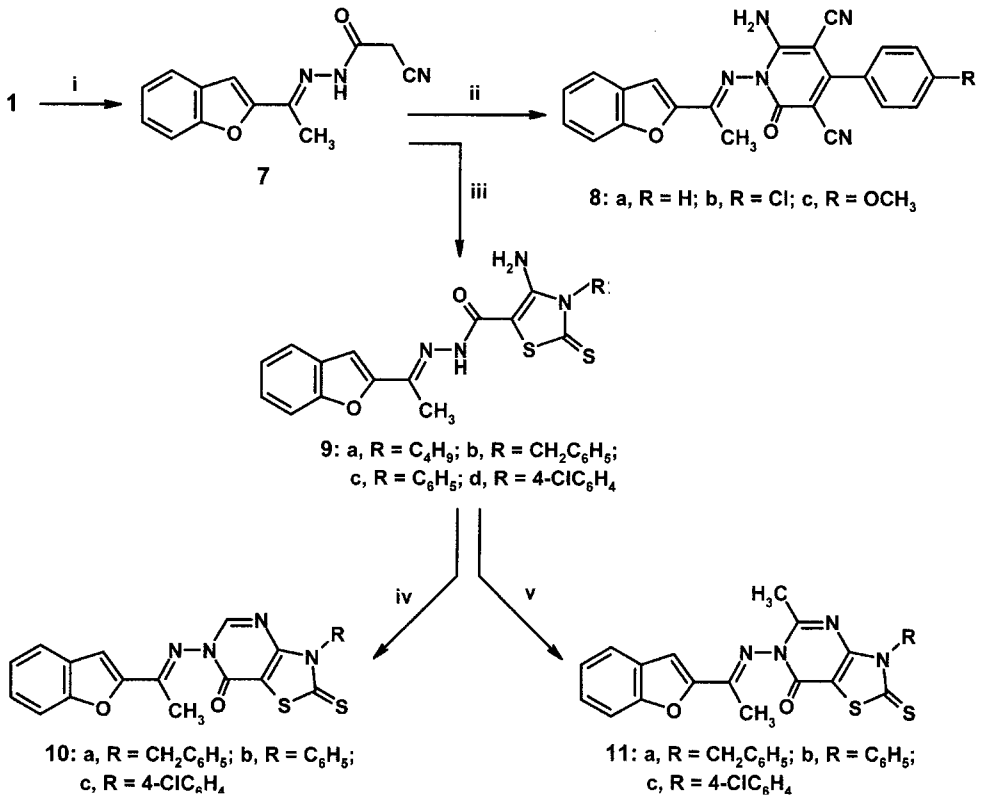
The target compounds contained the benzofuran backbone of the selected structure leads I, II, and III (Fig. 1), which were linked to the aforementioned bioactive heterocyclic ring systems at the 2-position through a two to four atoms spacer. The aim of these structural modifications was to obtain new non-nucleoside or antimetabolite hybrid structures with selective and synergistic chemotherapeutic efficacy.

The synthetic approach was confined to two novel series. The first series was derived from 1-(1-benzofuran-2-yl-ethylidine)-4-substituted-thiosemicarbazides (compounds **2a-d**, Scheme 1). This included the substituted thiazole and thiazolidinone moieties linked to benzofuran with a three atoms spacer (Scheme 1). The second series was derived from cyanoacetic acid(1-benzofuran-2-yl-ethylidene) hydrazide (**7**). Its members consisted of substituted 2-pyridinone, thiazole and thiazolopyrimidine moieties attached to benzofuran by a 2, 4 and 2 atoms spacer, respectively (Scheme 2).




**Reagents:** i = R-NH-CS-NHNH$_2$/ HOC$_2$H$_5$; ii = CH$_3$COCH$_2$Cl/dry dioxane; iii = 4-R$^1$C$_6$H$_4$COCH$_2$Br/dry dioxane; iv = ClCH$_2$CO$_2$H/anhd. CH$_3$CO$_2$Na, glacial AcOH; v = 4-R$^1$C$_6$H$_4$CHO / piperidine, dry dioxane.

**Scheme 1.** Synthetic scheme of benzofuran thiosemicarbazide derivatives




**Scheme 2.** Synthetic scheme of benzofuran thiazolidione derivatives

## MATERIAL AND METHODS

### Chemistry

All the melting points were determined in open-glass capillaries on a Gallenkamp melting point apparatus and were uncorrected. The IR spectra were recorded using KBr discs on a Perkin-Elmer 1430 spectrophotometer. $^1$H-NMR spectra were recorded in $CDCl_3$ on a Varian Gemini (90 or 400 MHz) and Jeol (400 or 500 MHz) spectrometers using TMS as the internal standard (chemical shift in δ ppm). $^{13}$C-NMR spectra were performed on Varian Gemini (100 MHz) spectrometer. The mass spectra (MS) were run on a Finnigan mass spectrometer, model SSQ/7000 (70 ev.). The microanalyses were performed at the microanalytical laboratory, National Research Center, Cairo, and the data was within ± 0.4% of the theoretical values. The reactions were monitored by thin-layer chromatography on silica gel-protected aluminium sheets (Type 60 F254, Merck), and the spots were detected by exposing the sheets to UV radiation at 254 nm for a few seconds using a UV-lamp.

#### 1-(1-Benzofuran-2-yl-ethylidene)-4-substituted-thiosemicarbazides (2a-d)

A mixture containing 2-acetyl benzofuran (**1**) (0.8 gm, 0.005 mol) and the appropriate 4-substituted thiosemicarbazide (0.005 mol) in absolute ethanol (30 mL) was heated under reflux for 3-4 h, and then cooled down to room temperature. The separated crystals were filtered, air-dried and recrystallized from chloroform. IR for **2a-d** (KBr, $cm^{-1}$): 3346-3278, 3249-3191 (N-H), 1601-1586 (C=N), 1534-1516, 1502-1488 (C=C, δ NH), 1569-1563, 1260-1257, 1188-1175, 936-926 (N-C=S amide I, II, III, and IV bands), 1250-1243, 1037-1019 (C-O-C); $^1$H-NMR for **2b** ($CDCl_3$, Varian Gemmini, 90 MHz, δ ppm); 2.2 (s, 3H, -N=C-C<u>H</u>$_3$), 4.85 (d, J = 7.0 Hz, 2H, -NH-C<u>H</u>$_2$-), 6.8 (s, 1H, benzofuran-$C_3$-<u>H</u>), 7.0-7.25 (m, 7H, phenyl and benzofuran-$C_5$-<u>H</u>, $C_6$-<u>H</u>), 7.3 (d, J = 4.5 Hz, 1H, benzofuran- $C_4$-<u>H</u>), 7.4 (d, J = 4.5 Hz, 1H, benzofuran-$C_7$-<u>H</u>), 7.85 (s, 1H, -N<u>H</u>-$CH_2$-, $D_2O$ exchangeable), 8.55 (s, 1H, C=N-N-<u>H</u>, $D_2O$ exchangeable); $^{13}$C-NMR for **2b** ($CDCl_3$, Varian Gemmini, 90 MHz, δ ppm): 12.7 ($CH_3$), 48.3 ($CH_2$), 107, 111, 121, 123, 124, 127.9, 152.6, 155.3 ($C_{3, 7, 4, 5, 6, 3a, 2, 7a}$ of benzofuran ring), 126, 127, 128.7, 138.2, ($C_4$, $C_{2/6}$, $C_{3/5}$, $C_1$ of phenyl ring), 149.8 (C=N), 178 (C=S); $^1$H-NMR for **2d** ($CDCl_3$, Varian Gemmini, 90 MHz, ä ppm): 2.35 (s, 3H, -$C_6H_4$-C<u>H</u>$_3$), 2.4 (s, 3H, -N=C-C<u>H</u>$_3$), 6.95 (s, 1H, benzofuran- $C_3$-<u>H</u>), 7.0-7.3 (m, 6H, phenyl and benzofuran-$C_5$-<u>H</u>, $C_6$-<u>H</u>), 7.35 (d, J = 4.5 Hz, 1H, benzofuran-$C_4$-<u>H</u>), 7.4 (d, J = 4.5 Hz, 1H, benzofuran-

$C_7$-H), 8.6 (s, 1H, -NH-$CH_2$-, $D_2O$ exchangeable), 9.2 (s, 1H, C=N-N-H, $D_2O$ exchangeable).

**_N_-(1-benzofuran-2-yl-ethylidene)-_N'_-(4-methyl-3-aryl-3_H_-thiazol-3-ylidene)-hydrazines (3a-c)**

A mixture containing the selected compounds **2a, b** or **2d** (0.002 mol) and chloroacetone (0.185 gm, 0.16 mL, 0.002 mol) in dry dioxane (10 mL) was refluxed for 2-3 h, cooled down to room temperature and diluted with few drops of water. The separated crystals were filtered, air-dried and recrystallized from aqueous ethanol. IR for **3a-c** (KBr, $cm^{-1}$): 1610-1608 (C=N), 1589-1580, 1551-1550, 1513-1492 (C=C), 1256, 1063-1049 (C-O-C); $^1$H-NMR for **3a** ($CDCl_3$, Varian Gemmini, 90 MHz, δ ppm): 2.0 (s, 3H, thiazole-$CH_3$), 2.3 (s, 3H, -N=C-$CH_3$), 5.6 (s, 2H, N-$CH_2$-), 5.6 (s, 1H, thiazole-$C_5$-H), 6.85 (s, 1H, benzofuran-$C_3$-H), 7.0-7.25 (m, 7H, phenyl and benzofuran-$C_5$-H, $C_6$-H), 7.3 (d, J = 4.5 Hz, 1H, benzofuran-$C_4$-H), 7.4 (d, J = 4.5 Hz, 1H, benzofuran-$C_7$-H); $^1$H-NMR for **3b** ($CDCl_3$, Varian Gemmini, 90 MHz, δ ppm): 1.8 (s, 3H, thiazole-$CH_3$), 2.1 (s, 3H, -N=C-$CH_3$), 5.7 (s, 1H, thiazole-$C_5$-H), 6.75 (s, 1H, benzofuran-$C_3$-H), 6.9-7.2 (m, 7H, phenyl and benzofuran-$C_5$-H, $C_6$-H), 7.25 (d, J = 4.5 Hz, 1H, benzofuran-$C_4$-H), 7.3 (d, J = 4.5 Hz, 1H, benzofuran-$C_7$-H).

**_N_-(1-benzofuran-2-yl-ethylidene)-_N'_-(3,4-diaryl-3_H_-thiazol-ylidene)-hydrazines (4a-f)**

A mixture containing the selected compounds **2b,c** or **2d** (0.002 mol) and the appropriate phenacyl bromide (0.002 mol) in dry dioxane (10 mL) was refluxed for 1-2 h. After cooling down to room temperature, the separated crystals were filtered, air dried and recrystallized from aqueous ethanol. IR for **4a-f** (KBr, $cm^{-1}$): 1609-1596, 1587-1584, 1563-1550, 1518-1513, 1505-1500 (C=N, C=C), 1257-1238, 1064-1036 (C-O-C); $^1$H-NMR for **4b** ($CDCl_3$, Varian Gemmini, 90 MHz, δ ppm): 2.25 (s, 3H, -$C_6H_4$-$CH_3$), 2.3 (s, 3H, -N=C-$CH_3$), 4.9 (s, 2H, N-$CH_2$-), 5.75 (s, 1H, thiazole-$C_5$-H), 6.8 (s, 1H, benzofuran-$C_3$-H), 6.9-7.25 (m, 11H, phenyl, p-methylphenyl and benzofuran-$C_5$-H, $C_6$-H), 7.3 (d, J = 4.5 Hz, 1H, benzofuran-$C_4$-H), 7.35 (d, J = 4.5 Hz, 1H, benzofuran-$C_7$-H); $^1$H-NMR for **4c** ($CDCl_3$, Varian Gemmini, 400 MHz, δ ppm): 2.2 (s, 3H, -N=C-$CH_3$), 6.2 (s, 1H, thiazole-$C_5$-H), 7.05 (s, 1H, benzofuran-$C_3$-H), 7.07-7.28 (m, 7H, phenyl and benzofuran-$C_5$-H, $C_6$-H), 7.3 (d, J = 8 Hz, 1H, benzofuran-$C_4$-H), 7.35 (d, J = 8 Hz, 1H, benzofuran-$C_7$-H), 7.53, 7.57 (dd, J = 7 Hz, 4H, 4-chlorophenyl); $^{13}$C-NMR for **4c** ($CDCl_3$ Varian Gemmini, δ ppm): 14 ($CH_3$), 105.6, 111.6, 121.1, 122.8, 124.9, 128.3, 153, 155.2 ($C_{3,7,4,5,6,3a,2,7a}$ of benzofuran ring), 102, 135, 169 ($C_{5,4,2}$ of thiazole ring), 116.5, 125.7, 129.3, 138 ($C_{2/6,4,3/5,1}$ of phenyl ring), 127.6, 128.8, 130, 134 ($C_{2/6,3/5,4,1}$ of p-chlorophenyl ring), 148 (C=N).

**2-[(1-Benzofuran-2-yl-ethylidene)hydrazono]-3-substituted-thiazolidin-4-ones (5a-d)**

A mixture containing the selected compounds **2a-d** (0.002 mol), monochloroacetic acid (0.28 gm, 003 mol) and anhydrous sodium acetate (0.35 gm, 0.005 mol) in glacial acetic acid (20 mL) was refluxed for 4-5 h, and then poured onto crushed ice. The formed precipitate was filtered, washed with water and crystallized from acetone. IR for **5a-d** (KBr, $cm^{-1}$): 1726-1695 (C=O), 1646-1644, 1609-1596 (C=N), 1568-1560, 1549-1530, 1506-1490 (C=C), 1261-1254, 1063-1044 (C-O-C); $^1$H-NMR for **5a** ($CDCl_3$, Varian Gemmini, 90 MHz, δ ppm): 1.0 (t, J = 7 Hz, 3H, -$CH_2$-$CH_3$), 1.5 (m, 4H, -$CH_2$-$CH_2$-$CH_3$), 2.4 (s, 3H, -N=C-$CH_3$), 3.7 (s, 2H, -S-$CH_2$-C=O), 3.8 (t, J = 7 Hz, 2H, N-$CH_2$-$CH_2$-), 7.0 (s, 1H, benzofuran-$C_3$-H), 7.1-7.3 (m, 2H, benzofuran-$C_5$-H, $C_6$-H), 7.35 (d, J = 4.5 Hz, 1H, benzofuran-$C_4$-H), 7.4 (d, J = 4.5 Hz, 1H, benzofuran-$C_7$-H); $^{13}$C-NMR for **5a** ($CDCl_3$, Varian Gemmini, δ ppm): 13.7 ($CH_3$), 14.4, 20.2, 29.12, 43.4 ($C_{4,3,2,1}$ of butyl group), 32.5, 162.7, 172 ($C_{5,2,4}$ of thiazole ring), 107.7, 111.6, 121, 123.19, 126.3, 128.1, 153.8, 155.4 ($C_{3,7,4,5,6,3a,2,7a}$ of benzofuran ring), 154.8 (C=N); $^1$H-NMR for **5b** ($CDCl_3$, Varian Gemmini, 90 MHz, δ ppm): 2.4 (s, 3H, -N=C-$CH_3$), 3.7 (s, 2H, S-$CH_2$-C=O), 4.9 (s, 2H, N-$CH_2$-phenyl), 7.0 (s, 1H, benzofuran-$C_3$-H), 7.1-7.3 (m, 7H, phenyl and benzofuran-$C_5$-H, $C_6$-H), 7.35 (d, J = 4.5 Hz, 1H, benzofuran-$C_4$-H), 7.4 (d, J = 4.5 Hz, 1H, benzofuran-$C_7$-H); $^{13}$C-NMR for **5b** ($CDCl_3$, Varian Gemmini, δ ppm ): 13.5 ($CH_3$), 24.4 (N-$CH_2$-phenyl), 113, 114,139, 125, 126, 128, 129, 150 ($C_{3,7,4,5,6,3a,2,7a}$ of benzofuran ring), 30, 153, 165 ($C_{5,2,4}$ of thiazole ring), 138, 128.3, 129, 125 ($C_{4,2/6,3/5,1}$ of phenyl ring), 140 (C=N); $^1$H-NMR for **5c** ($CDCl_3$, Varian Gemmini, 90 MHz, δ ppm): 2.25 (s, 3H, -N=C-$CH_3$), 3.9 (s, 2H, S-$CH_2$-C=O), 7.0 (s, 1H, benzofuran-$C_3$-H), 7.1-7.35 (m, 7H, phenyl and benzofuran-$C_5$-H, $C_6$-H), 7.4 (d, J = 4.5 Hz, 1H, benzofuran-$C_4$-H), 7.45 (d, J = 4.5 Hz, 1H, benzofuran-$C_7$-H); $^{13}$C-NMR for **5c** ($CDCl_3$, Varian Gemmini, δ ppm): 14.5 ($CH_3$), 108, 111.8, 121.6, 123.2, 125.9, 128.8, 153, 155.4 ($C_{3,7,4,5,6,3a,2,7a}$ of benzofuran ring), 32, 163, 172 ($C_{5,2,4}$ of thiazole ring), 127.6, 128, 129, 134 ($C_{2,4,3,1}$ of phenyl ring), 156 (C=N).

Single-crystal X-ray analysis of compound **5c**: The diffraction data of $C_{19}H_{15}N_3O_2S$ (mol. wt. 349.41), were collected from cubic colorless crystals of dimensions 0.21 $mm^{-1}$ using an automatic diffractometer maXus (Bruker Nonius, Delft & McScience, Japan). The crystal system was found to be monochromic with a = 23.7452(11) Å, b = 5.5809(3) Å, c = 12.8692(6) Å, α = 90.00 Å, β = 101.926 (3) Å, γ = 90.00 Å, V = 1668.61(14) Å, Z = 4, $D_{calc}$ = 1.3339 $g/cm^3$. Diffraction data: 1195 reflections were collected using Moka radiation (λ = 0.71073). Computing data collection: Kappa CCD. The structure was solved by the direct method (Altomar *et al.,* 1994) using the program, SHELXS-97. The agreement factor was R = 0.056.

**2-[(1-Benzofuran-2-yl-ethylidene)hydrazono]-5-(4-substituted benzylidene)-3-substituted-thiazolidin-4-ones (6a-d)**

A mixture containing compound **5b** or **5c** (0.002 mol), 4-substituted benzaldehyde derivative (0.002 mol) and piperidine (2 drops) in dry dioxane (10 mL) was refluxed for 3-4 h, cooled down to room temperature. The separated crystals were filtered, air dried and recrystallized from aqueous ethanol. IR for **6a-d** (KBr, $cm^{-1}$): 1729-1697 (C=O), 1646-1644, 1617-1608 (C=N), 1597-1592, 1559-1550, 1520-1502 (C=C), 1259-1238, 1052-1030 (C-O-C); $^1$H-NMR for **6a** ($CDCl_3$, Jeol, 500 MHz, δ ppm): 2.5 (s, 3H, N=C-C$\underline{H}_3$), 5.17 (s, 2H, N-C$\underline{H}_2$-), 7.2-7.5 (m, 11H, phenyl, *p*-chlorophenyl and benzofuran-$C_5$-$\underline{H}$, $C_6$-$\underline{H}$), 7.58 (d, J = 7 Hz, 1H, benzofuran-$C_4$-$\underline{H}$), 7.62 (d, J = 7 Hz, 1H, benzofuran-$C_7$-$\underline{H}$), 7.7 (s, 1H, S-C=C-$\underline{H}$); $^1$H-NMR for **6d** ($CDCl_3$, Jeol, 400 MHz, δ ppm): 2.31 (s, 3H, N=C-C$\underline{H}_3$), 3.89 (s, 3H, -OC$\underline{H}_3$), 7.0 (s, 1H, benzofuran-$C_3$-$\underline{H}$), 7.14-7.46 (m, 7H, benzofuran-$C_5$-$\underline{H}$, $C_6$-$\underline{H}$ and phenyl), 7.48 (d, J = 7Hz, 1H, benzofuran-$C_4$-$\underline{H}$), 7.53 (d, J = 7Hz, 1H, benzofuran-$C_7$-$\underline{H}$), 7.62 (2d, J = 4 Hz, 4H, p-methoxyphenyl), 7.81 (s, 1H, S-C=C-$\underline{H}$); $^{13}$C-NMR for **6d** ($CDCl_3$, Varian Gemmini, δ ppm): 15.1 ($CH_3$), 55.8 ($OCH_3$), 108.6, 112.3, 121, 122, 123.6, 128.5, 156, 156.1 ($C_{3,7,4,5,6,3a,2,7a}$ of benzofuran ring), 115.04, 129.4, 139, 153 ($C_{3/5,2/6,1,4}$ of *p*-methoxyphenyl ring), 127, 129, 128.1, 135 ($C_{2/6,4,3/5,1}$ of phenyl ring), 132, 161.3, 178 ($C_{5,2,4}$ of thiazole ring), 135 (S-C=C-H), 154 (C=N).

**6-Amino-1-(1-benzofuran-2-yl-ethylideneamino)-2-oxo-4-aryl-1,2-dihydro-pyridine-3,5-dicarbonitrile (8a-c)**

A mixture containing compound **7** (0.48 gm, 0.002 mol), the appropriate 2-arylidenemalononitrile (0.002 mol) and 2 drops piperidine in absolute ethanol (20 mL) was refluxed for 2-3 h. The mixture was then cooled down to room temperature and diluted with few drops of water. The crystals formed were filtered, air dried and recrystallized from aqueous DMF. IR for **8a-c** (KBr, $cm^{-1}$): 3463-3461, 3357-3352 (N-H), 2217-2213 (CN), 1668-1660 (C=O), 1645-1638 (C=N), 1610-1581, 1552-1550, 1518-1516, 1501-1500 (C=C, δ NH), 1259-1248, 1074 (C-O-C); $^1$H-NMR for **8a** ($CDCl_3$, Jeol, 400 MHz, δ ppm): 1.25 (s, 1H, tautomeric =C-O$\underline{H}$), 2.1 (s, 3H, N=C-C$\underline{H}_3$), 5.42 (s, 2H, tautomeric N$\underline{H}_2$), 7.2 (s, 1H, benzofuran-$C_3$-$\underline{H}$), 7.28-7.42 (m, 2H, benzofuran-$C_5$-$\underline{H}$, $C_6$-$\underline{H}$), 7.5-7.57 (m, 5H, phenyl), 7.62 (d, J = 8 Hz, 1H, benzofuran-$C_4$-$\underline{H}$), 7.7 (d, J = 8.4 Hz, 1H, benzofuran-$C_7$-$\underline{H}$), 7.83 (s, 1H, tautomeric =N-$\underline{H}$); $^1$H-NMR for **8b** ($CDCl_3$, Jeol, 400 MHz, δ ppm): 1.25 (s, 1H, tautomeric =C-O$\underline{H}$), 2.1 (s, 3H, N=C-C$\underline{H}_3$), 5.43 (s, 2H, tautomeric-$NH_2$), 7.26 (s, 1H, benzofuran-$C_3$-$\underline{H}$), 7.29-7.43 (m, 2H, benzofuran-$C_5$-$\underline{H}$, $C_6$-$\underline{H}$), 7.48-7.56 (2d, 4H, 4-chlorophenyl), 7.59 (d, J = 8 Hz, 1H, benzofuran-$C_4$-$\underline{H}$), 7.69 (d, J = 8.4 Hz, 1H, benzofuran-$C_7$-$\underline{H}$), 7.85 (s, 1H, tautomeric =N-$\underline{H}$).

The electron impact Mass Spectrum for **8b** m/z (relative abundance %): $M^+$ (was not detected), 369[M-58, (100)], 371 (36.7), 304 (5.3), 251 (5.8), 125 (8).

**4-amino-3-substituted-2-thioxo-2,3-dihydro-thiazole-5-carboxylic acid-(1-benzofuran-2-yl-ethylidene)-hydrazides (9a-d)**

The appropriate isothiocyanate (0.005 mol) was added to a stirred solution of compound **7** (1.2 gm, 0.005 mol), finely divided sulfur (0.16 gm, 0.005 mol) and triethylamine (0.6 mL) in a dimethylformamide/ethanol (1:1, 20 mL) solvent mixture. The reaction mixture was heated under reflux for 1 h, during which time the product crystallized. The reaction mixture was allowed to cool down to room temperature, and the resulting product was filtered, washed with ethanol, air-dried and recrystallized from aqueous dimethylformamide. IR for **9a-d** (KBr, $cm^{-1}$): 3391-3347, 3279-3250, 3164-3156 (NH), 1663-1660 (C=O), 1628-1623 (C=N), 1601-1597, 1536-1534, 1513-1494 (C=C, δ NH), 1258-1238 (C=S), 1256-1248, 1060-1041 (C-O-C); $^1$H-NMR for **9a** ($CDCl_3$, Jeol, 400 MHz, δ ppm): 1.0 (t, J = 8 Hz, 3H, -$CH_2$-C$\underline{H}_3$), 1.6 (m, 4H, -C$\underline{H}_2$-C$\underline{H}_2$-$CH_3$), 2.3 (s, 3H, N=C-C$\underline{H}_3$), 4.2 (t, J = 8 Hz, 2H, N-C$\underline{H}_2$-$CH_2$-), 6.63 (br.s, 2H, N$\underline{H}_2$), 7.24 (s, 1H, benzofuran-$C_3$-$\underline{H}$), 7.28-7.44 (m, 2H, benzofuran-$C_5$-$\underline{H}$, $C_6$-$\underline{H}$), 7.56 (d, J = 8 Hz, 1H, benzofuran-$C_4$-$\underline{H}$), 7.65 (d, J = 8.4 Hz, 1H, benzofuran-$C_7$-$\underline{H}$), 8.5 (br.s, 1H, -$\underline{H}$N-C=O).

**6-(1-Benzofuran-2-yl-ethylideneamino)-3-substituted-2-thioxo-2,3-dihydro-6H-thiazolo[4,5-d]-pyrimidin-7-one (10a-c)**

A solution containing the appropriate compound (**9b-d**) (0.002 mol) in a mixture of triethylorthoformate (10 mL) and acetic anhydride (10 mL) was heated under reflux for 3 h. During this time, the product partially crystallized. The reaction mixture was allowed to cool down to room temperature. The separated product was filtered, washed with ethanol, air dried and recrystallized from aqueous dimethylformamide. IR for **10a-c** (KBr, $cm^{-1}$): 1690-1674 (C=O), 1647-1645, 1630-1626 (C=N), 1590, 1517-1515, 1492-1490 (C=C), 1265-1234 (C=S), 1250-1248, 1060-1050 (C-O-C); $^1$H-NMR for 10a ($CDCl_3$, Jeol, 500 MHz, δ ppm): 2.3 (s, 3H, N=C-C$\underline{H}_3$), 5.6 (s, 2H, N-C$\underline{H}_2$-), 7.2 (s, 1H, benzofuran-$C_3$-$\underline{H}$), 7.34-7.48 (m, 2H, benzofuran-$C_5$-$\underline{H}$, $C_6$-$\underline{H}$), 7.52-7.55 (m, 5H, phenyl), 7.6 (d, J = 8 Hz, 1H, benzofuran-$C_4$-$\underline{H}$), 7.69 (d, J = 8.4 Hz, 1H, benzofuran-$C_7$-$\underline{H}$), 8.2 (s, 1H, N-C$\underline{H}$=N-).

The electron impact Mass Spectrum for **10c** m/z (relative abundance %): 452 [$M^+$ (7.8)], 454 [$M^+$+2 (4.5)], 115 (100), 294 (1.3), 158 (58), 144 (54), 143 (32.9).

**6-(1-Benzofuran-2-yl-ethylideneamino)-5-methyl-3-substituted-2-thioxo-2,3-dihydro-6H-thiazolo[4,5-d]pyrimidin-7-ones (11a-c)**

A solution containing the appropriate compound (**9b-d**) (0.002 mol) in acetic anhydride (20 mL) was heated under reflux for 4-5 h. The reaction mixture was allowed to cooll down to room temperature, and the separated crystals were filtered, washed with ethanol, air-dried and recrystallized from dimethylformamide. IR for **11a-c** (KBr, $cm^{-1}$): 1677-1674 (C=O), 1645-1637 (C=N), 1599-1594, 1535-1519, 1493-1492 (C=C), 1260-1232 (C=S), 1245-1243, 1065-1052 (C-O-C); $^1$H-NMR for **11b** ($CDCl_3$, Jeol, 400 MHz, $\delta$ ppm): 2.3 (s, 3H, N=C-C$\underline{H}_3$), 2.5 (s, 3H, N=C(C$\underline{H}_3$)-N-), 7.28 (s, 1H, benzofuran-$C_3$-$\underline{H}$), 7.32-7.4 (m, 2H, benzofuran-$C_5$-$\underline{H}$, $C_6$-$\underline{H}$), 7.53-7.57 (m, 5H, phenyl), 7.6 (d, J = 7 Hz, 1H, benzofuran-$C_4$-$\underline{H}$), 7.68 (d, J = 7 Hz, 1H, benzofuran-$C_7$-$\underline{H}$); $^{13}$C-NMR for **11b** ($CDCl_3$, Varian Gemmini, $\delta$ ppm): 14.1 ($CH_3$), 21.1 (N=C($\underline{C}H_3$)-N-), 107, 111.7, 121, 123, 125, 128.2, 153, 155 ($C_{3,7,4,5,6,3a,2,7a}$ of benzofuran ring), 128, 129, 130, 135 ($C_{4,2/6,3/5,1}$ of phenyl ring), 148 (C=N), 149, 156, 157, 165, 167 ($C_{7a,5,3a,7,2}$ of thiazolopyrimidine ring).

## Biological evaluation

***In vitro*** anti-HIV activity

The *in vitro* anti-HIV drug testing system was performed in the national Cancer Institute's Developmental Therapeutics Program, AIDS antiviral screening program, according to a procedure reported in the literature (Weislow *et al.,* 1989). The assay involved the killing of $T_4$ lymphocytes by HIV. The $T_4$ lymphocytes (CEM cell line) were exposed to HIV at a virus-to-cell ratio of approximately 0.05 and treated with the test compounds dissolved in dimethylformamide, at doses ranging from $10^{-8}$ to $10^{-4}$ M. A complete virus reproduction cycle is essential to obtain the required level of cell death (incubation at 37°C in a 5% carbon dioxide atmosphere for 6 days). The uninfected cells exposed to the compound were used as a toxicity control, whereas the infected and uninfected cells without the compound were used as the basic controls. After incubation, the tetrazolium salt, XTT, was added to the wells and incubated to allow formazan color development by the viable cells. The level of formazan production was measured specrophotometrically and any potential protective activity was confirmed by the microscopic detection of viable cells. The effect of each compound on the cell growth of HIV-infected and uninfected cells was compared with that of the untreated uninfected cells. All the tests were compared with AZT as a positive control carried out at the same time under identical conditions.

**Antineoplastic activity**

Compounds **4a,c**, **6a-d**, **10a-c**, and **11a-c** were evaluated in three cell lines (Lung NCI-H460, Breast MCF 7 and CNS SF-268) using a one dose primary anticancer assay following a NCI preclinical antitumor drug discovery screen (Grever *et al.,* 1992).

In the current protocol, each cell was inoculated and preincubated on a microtiter plate. The test agents were then added at a single concentration (100 μM), and the culture was incubated for 48 h. The endpoint was determined using alamar blue. The results for each agent are presented as the percentage of the growth of the treated cells compared with that of the untreated control cells.

**Antimicrobial activity**

The agar diffusion technique (Conte *et al.,* 1988) was used to evaluate the tested compounds using a 2 mg/mL solution in DMF. The test organisms were *Staphylococcus aureus* (ATCC 6538) and *bacillus subtilis* (DB 100) as Gram-positive bacteria, *Pseudomonas aeruginosa* (ATTC 27853) and *Escherichia coli* (DH5a) as Gram-negative bacteria and *Candida albicans* (0443P) as a representative fungi. A control using DMF without the test compound was included for each organism. The minimal inhibitory concentration (MIC) of the most active compounds was measured using a two-fold serial broth dilution method (Scott, 1989). Ampicillin, and clotrimazol in DMF were used as the reference drugs.

# RESULTS AND DISCUSSION

## Chemistry

Schemes 1 and 2 show the synthetic strategies used to obtain the target compounds. Reacting 2-acetybenzofuran (**1**) with the appropriate 4-substituted thiomemicarbazide afforded the respective key intermediate thiosemicarbazones (**2a-d**). Cyclocondensation of the latter with either chloroacetone or the appropriate substituted phenacyl bromide yielded the *N*-(1-benzofuran-2-yl-ethylidene)-*N'*-(3,4-disubstituted-3*H*-thiazol-2-ylidene)-hydrazines (**3a-c** and **4a-f**).

Cyclization of compounds **2a-d** with chloroacetic acid gave 2-[(1-benzofuran-2-yl-ethylidene)hydrazono]-3-substituted-thiazolidin-4-ones (**5a-d**). Condensation of the latter with the aromatic aldehydes in the presence of piperidine yielded the corresponding 3,5-disubstituted-thiazolidin-4-ones (**6a-d**).

Scheme 2 starts with the key intermediate, cyanoacetic acid-(1-benzofuran-2-yl-ethylidene)-hydrazide (**7**), which was prepared using a procedure described elsewhere (Villere *et al.,* 1958). The target 6-amino-4-aryl-1-(1-benzofuran-2-yl-ethylideneamino)-2-oxo-1,2-dihydro-pyridine-3,5-dicarbonitriles (**8a-c**) were obtained by reacting compound **7** with the appropriate 2-arylidenemalononitrile according to the reaction conditions reported for the synthesis

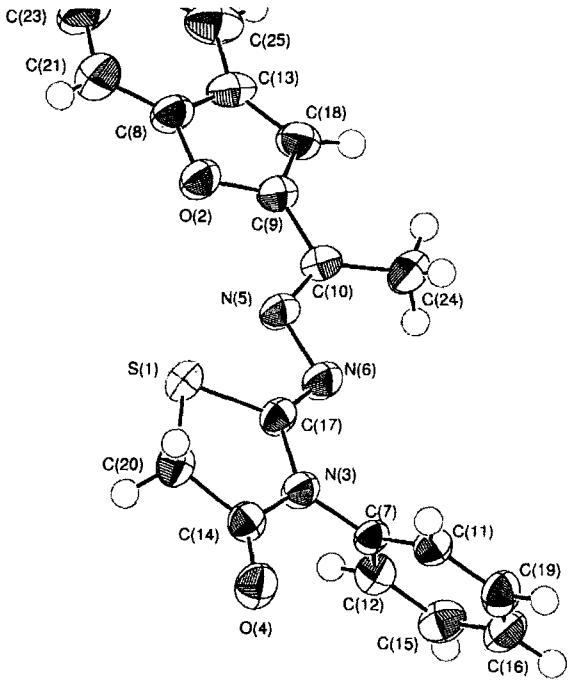



**Fig. 2.** Structure and solid state absolute configuration of compound **5c**

of its related compounds (Rida *et al.*, 1995). On the other hand, the synthesis of 4-amino-3-substituted-2-thioxo-2,3-dihydro-thiazole-5-carboxylic acid-(1-benzofuran-2-yl-ethylidene)-hydrazides (**9a-d**) was achieved using a procedure reported for preparing analogous compounds (Badaway *et al.*, 1992). This procedure involved reacting compound **7** with sulfur and the selected arylisothiocyanate in presence of a catalytic amount of piperidine. Cyclization of compounds **9a-d** with either acetic anhydride or with a mixture of acetic anhydride and triethyl orthoformate gave the corresponding 6-(1-benzofuran-2-yl-ethylideneamino)-3-substituted-2-thioxo-2,3-dihydro-6*H*-thiazolo-[4,5-*d*]-pyrimidin-7-ones (compounds **10a-c** and **11a-c** respectively).

The structures of the newly synthesized compounds were confirmed by microanalyses, as well as by the IR and $^1$H-NMR spectral data. For further confirmation, some selected compounds were also examined by $^{13}$C-NMR and electron impact spectroscopy.

The availability of the X-ray crystallographic analysis clarified the absolute configuration of compound **5c** (Fig. 2). The benzofuran ring was confirmed to be located in an antiposition to the 3-phenyl-4-oxothiazolidin-2-ylidenamino moiety. The phenyl group on the thiazolidine nitrogen is non-coplanar and exists mainly in a plane perpendicular to the thiazolidine ring.

## Biological study

### *In vitro* anti-HIV-1 activity

Nine compounds (**5**, **6a,b**, **7a-c**, and **11a-c**) were selected by NCI and evaluated for their potency against HIV-1 induced cytopathogenicity in a human $TB_{4B}$ lymphocyte cell line (CEM). The procedure used was designed to detect the agents acting at any stage of the virus reproductive cycle (Weislow *et al.*, 1989). The assay involved the killing of $T_4$ lymphocytes by HIV-1. Compounds that interact with the virons or virus gene-product to interfere with the viral activities will protect the cells from cytolysis. The activity is expressed as % protection, which indicates the percentage of surviving HIV-infected cells treated with the test compounds (at the indicated concentration) relative to the same uninfected untreated controls. The median effective concentration ($EC_{50}$) of the test compounds using the infected cells was compared with their cytotoxic effect ($IC_{50}$) in the uninfected cultures. The $EC_{50}$ indicates the concentration of the test agent resulting in a 50% reduction of the viral cytopathic effect. The $IC_{50}$ represents the toxic concentration of the drug that inhibits the growth of normal uninfected cells by 50%. The therapeutic index ($TI_{50}$) was determined by dividing the ($IC_{50}$) by ($EC_{50}$). The infected and uninfected cultures incubated without the test compounds were used as the controls. The zidovudine (AZT) treated cultures were also used as a positive control.

Among the tested compounds, 4-amino-3-butyl-2-thioxo-2,3-dihydro-thiazole-5-carboxylic acid-(1-benzofuran-2-yl-ethylidene)-hydrazide (**9a**) was confirmed to have moderate *in vitro* anti-HIV-1 activity. This compound at a concentration of $2.5\times10^{-5}$ M reduced the viral cytopathic effect at levels ranging from 54.7%-59.55% in four independent experiments. On the other hand, compound 2-[(1-benzofuran-2-yl-ethylidene)hydrazono]-3-phenyl-thiazolidin-4-one (**5c**) had a larger antiviral cytopathic effect (93.19% and 88.94%) at a concentration $2.0\times10^{-4}$ M in two independent experiments. To confirm the activity the assay experiments were repeated after one and two months, which showed a significant decrease in activity. Accordingly, the compound was graded as "inactive" by the NCI. It is unclear if the declining antiviral activity is the result of compound decay, or the initial results did not reflect the true activity.

Compound **2a** showed weak activity (46.34% reduction of viral cytopathic effects). The remaining compounds failed to show an anticytopathic effect on HIV because the cell growth of the HIV-infected cells remained was within 7-19%. The recorded $IC_{50}$% and $EC_{50}$% for compounds **5c** and **9a** were (> $2.00\times10^{-4}$ M, $2.56\times10^{-5}$ M and $9.85\times10^{-5}$ M, $4.83\times10^{-6}$ M respectively). The therapeutic indices (TI) were > 2.03 and 5.29, which were insufficient to justify further *in vivo* testing compared with AZT (TI = > $3.89\times10^{-2}$, Table II).

### Anticancer screening

Anticancer screening was performed at the National

**Table I.** Physical and analytical data of the compounds synthesized

| Cpd. No. | R | $R^1$ | M.P. | Yield % | Mol. Formula (Mol. Wt.) |
|---|---|---|---|---|---|
| **2a** | $C_4H_9$ | - | 78-80 | 95 | $C_{15}H_{19}N_3OS$ (289.4) |
| **2b** | $CH_2C_6H_5$ | - | 166-8 | 90 | $C_{18}H_{17}N_3OS$ (323.4) |
| **2c** | $C_6H_5$ | - | 205-7 | 90 | $C_{17}H_{15}N_3OS$ (309.39) |
| **2d** | 4-$CH_3C_6H_4$ | - | 171-2 | 75 | $C_{18}H_{17}N_3OS$ (323.4) |
| **3a** | $CH_2C_6H_5$ | - | 119-21 | 67 | $C_{21}H_{19}N_3OS$ (361.47) |
| **3b** | $C_6H_5$ | - | 127-9 | 55 | $C_{20}H_{17}N_3OS$ (355.46) |
| **3c** | 4-$CH_3C_6H_4$ | - | 134-6 | 54 | $C_{21}H_{19}N_3OS$ (361.47) |
| **4a** | $CH_2C_6H_5$ | Cl | 157-9 | 60 | $C_{26}H_{20}ClN_3OS$ (485.05) |
| **4b** | $CH_2C_6H_5$ | $CH_3$ | 152-3 | 85 | $C_{27}H_{23}N_3OS$ (446.57) |
| **4c** | $C_6H_5$ | Cl | 225-6 | 68 | $C_{25}H_{18}ClN_3OS$ (443.95) |
| **4d** | $C_6H_5$ | $CH_3$ | 110-2 | 65 | $C_{26}H_{21}N_3OS$ (423.53) |
| **4e** | 4-$CH_3C_6H_4$ | Cl | 240-2 | 50 | $C_{26}H_{20}ClN_3OS$ (457.98) |
| **4f** | 4-$CH_3C_6H_4$ | $CH_3$ | 222-4 | 85 | $C_{27}H_{23}N_3OS$ (437.60) |
| **5a** | $C_4H_9$ | - | 127-128 | 76 | $C_{17}H_{19}N_3O_2S$ (347.42) |
| **5b** | $CH_2C_6H_5$ | - | 156-157 | 65 | $C_{20}H_{17}N_3O_2S$ (363.43) |
| **5c** | $C_6H_5$ | - | 235-236 | 85 | $C_{19}H_{15}N_3O_2S$ (349.41) |
| **5d** | 4-$CH_3C_6H_4$ | | 221-223 | 55 | $C_{20}H_{17}N_3O_2S$ (363.50) |
| **6a** | $CH_2C_6H_5$ | Cl | 225-227 | 60 | $C_{27}H_{20}ClN_3O_2S$ (485.99) |
| **6b** | $CH_2C_6H_5$ | $OCH_3$ | 137-139 | 76 | $C_{28}H_{23}N_3O_3S$ (499.58) |
| **6c** | $C_6H_5$ | Cl | 222-224 | 70 | $C_{26}H_{18}ClN_3O_2S$ (471.96) |
| **6d** | $C_6H_5$ | $OCH_3$ | 237-239 | 67 | $C_{27}H_{21}N_3O_3S$ (467.55) |
| **8a** | H | - | 246-248 | 57 | $C_{23}H_{15}N_5O_2$ (393.41) |
| **8b** | Cl | - | 303-305 | 35 | $C_{23}H_{14}ClN_5O_2$ (427.84) |
| **8c** | $OCH_3$ | - | 251-253 | 35 | $C_{24}H_{17}N_5O_3$ (423.43) |
| **9a** | - | $C_4H_9$ | 214-216 | 50 | $C_{18}H_{20}N_4O_2S_2$ (424.54) |
| **9b** | - | $CH_2C_6H_5$ | 226-228 | 55 | $C_{21}H_{18}N_4O_2S_2$ (422.53) |
| **9c** | - | $C_6H_5$ | 243-244 | 50 | $C_{20}H_{16}N_4O_2S_2$ (408.5) |
| **9d** | - | 4-$ClC_6H_4$ | 251-253 | 50 | $C_{20}H_{15}ClN_4O_2S_2$ (442.95) |
| **10a** | - | $CH_2C_6H_5$ | 273-274 | 70 | $C_{22}H_{16}N_4O_2S_2$ (432.52) |
| **10b** | - | $C_6H_5$ | 138-140 | 80 | $C_{21}H_{14}N_4O_2S_2$ (418.49) |
| **10c** | - | 4-$ClC_6H_4$ | 219-221 | 78 | $C_{21}H_{13}ClN_4O_2S_2$ (452.94) |
| **11a** | - | $CH_2C_6H_5$ | 240-241 | 95 | $C_{23}H_{18}N_4O_2S_2$ (446.55) |
| **11b** | - | $C_6H_5$ | 204-206 | 70 | $C_{22}H_{16}N_4O_2S_2$ (432.52) |
| **11c** | - | 4-$ClC_6H_4$ | 230-232 | 65 | $C_{22}H_{15}ClN_4O_2S_2$ (466.97) |

**Table II.** *In vitro* anti- HIV-1 activity of compounds **5c**, **9a**, and AZT

| Comp. No. | $IC_{50}$[a] (Molar) | $EC_{50}$[b] (Molar) | TI ($IC_{50}/EC_{50}$) |
|---|---|---|---|
| 5c | $>2.00\times10^{-4}$ | $9.8\times10^{-5}$ | >2.03 |
| 9a | $2.56\times10^{-5}$ | $4.83\times10^{-6}$ | 5.29 |
| AZT[c] | $>1.00\times10^{-6}$ | $2.57\times10^{-9}$ | $>3.89\times10^{-2}$ |

[a] 50% inhibitory concentration (molar concentration of the compounds that cause a 50% inhibition of cell growth).
[b] 50% effective concentration (molar concentration of compounds that cause 50% protection against the HIV-1 cytopathic effect).
[c] Positive control.

Cancer Institute (NCI), Bethesda, Maryland, U.S.A.. Twelve compounds were selected by NCI (**4a,c**, **6a-d**, **10a-c**, and **11a-c**) and evaluated for their *in vitro* antineoplastic activity. These compounds were tested using the lung NCI-H-460, Breast MCF-7 and CNS SF-268 cell lines, with a one dose primary anticancer assay (Grever *et al.*, 1995). The results are reported as the percentage growth of the treated cells compared with the untreated control cells (Table III). Compounds that reduce the growth of any one of the cell lines to 32% or less (negative numbers indicate cell death) were passed on for further evaluation using a full panel of 60 cell lines at five log dose ranges. Unfortunately, none of the tested compounds showed any activity. However, compounds **4a**, **10c**, and **11b** showed weak activity against the Breast MCF7 and Lung NCI-H460 cell lines. Nevertheless, further structural modification of these structural leads might lead to the production of new anticancer agents.

## Antimicrobial testing

All the synthesized compounds were preliminary evaluated for their *in vitro* activity against *S. aureus* as Gram-positive bacteria, *E. coli* as gram-negative bacteria and *C. albicans* as a representative fungi. Ampicillin and clotrimazole were used as the reference drugs.

Table IV shows that compounds **2a**, **2d**, and **5c** exhibited mild activity against *C. albicans* (MIC 125 mg/mL). While

**Table III.** Growth percentages of the 3-cell line panel in the primary anticancer screen of some selected compounds

| Comp. No | NSC NO | Sample Concentration | Percentage growth | | |
|---|---|---|---|---|---|
| | | | Breast-MCF7 | Lung NCI-H460 | CNS SF-268 |
| **4a** | S-725616-V | 1.00-04 M | 69 | 66 | 121 |
| **4c** | S-725615-U | 1.00-04 M | 102 | 123 | 142 |
| **6a** | S-725707-U | 1.00-04 M | 114 | 148 | 175 |
| **6b** | S-725705-S | 1.00-04 M | 108 | 100 | 155 |
| **6c** | S-725617-w | 1.00-04 M | 95 | 119 | 167 |
| **6d** | S-725706-T | 1.00-04 M | 114 | 140 | 160 |
| **10a** | S-725709-W | 1.00-04 M | 110 | 136 | 148 |
| **10b** | S-725708-V | 1.00-04 M | 118 | 142 | 159 |
| **10c** | S-725710-X | 1.00-04 M | 78 | 66 | 101 |
| **11a** | S-725712-Z | 1.00-04 M | 116 | 126 | 145 |
| **11b** | S-725711-Y | 1.00-04 M | 68 | 62 | 109 |
| **11c** | S-725701-O | 1.00-04 M | 90 | 109 | 124 |

**Table IV.** The inhibition zones (IZ) in mm diameter, and the minimal inhibitory concentration (MIC) in μg/mL of the active compounds

| Comp. No. | S. aureus | | E. coli | | C. albicans | |
|---|---|---|---|---|---|---|
| | IZ | MIC | IZ | MIC | IZ | MIC |
| 2a | 14 | 500 | -[a] | - | 14 | 125 |
| 2d | - | - | - | - | 22 | 125 |
| 3a | - | - | 18 | 500 | - | - |
| 5a | - | - | - | - | 12 | 500 |
| 5c | 14 | 500 | - | - | 14 | 125 |
| 11c | 12 | 500 | 14 | 500 | 12 | 500 |
| Ampicillin | - | 5 | - | 10 | - | - |
| Clotrimazole | - | - | - | - | - | 10 |

[a] Inactive, inhibition zone < 12 mm.

compounds **5a** and **11c** had lower antifugal activity (MIC 500 μg/mL). On the other hand, compounds **2a**, **5c**, and **11c** demonstrated weak activity against the Gram-positive *S. aureus* (MIC 500 μg/mL) while compound **3a** and **11c** showed weak activity against the Gram-negative *E. coli* (MIC 500 μg/mL).

## CONCLUSION

The anti-HIV, antitumor and antimicrobial screening data show that the substituted thiosemicarbazides (**2a-d**) have weak activity. Compound **2a** (R = n-butyl) offered 46% protection against the HIV-1 cytopathic effect, as well as mild antifungal and weak antibacterial activities while compound **2d** had a mild antifungal effect. The cyclization of compounds **2a-d** to the corresponding substituted thiazolidinon-2-ones (**5a-d**) increased the anti-HIV potency. Compound **5c** (R = phenyl) had the highest activity, which was decreased when R = *n*-butyl (**5a**) or when the phenyl group was separated by one carbon atom spacer in compound **5b**. Compound **5d** (R = p-tolyl) had no activity. Compounds **6a-d**, which are 5-alkylidene derivatives of **5a-d**, also had no activity. This suggests that substitution at the 5-position of the thiazole ring abolished its biological activity.

On the other hand, the cyclization of compounds **2a-d** afforded the substituted thiazole derivatives (compounds **3a-c**). Compound **3c** (R = p-tolyl) decreased the growth of breast and lung cancer to some extent compared with compound **3a** (R = benzyl), which was completely inactive. The thiazolo[4,5-d]pyrimidines, (**10c** and **11a**), showed weak activity against breast and lung cancer. It should be noted that the substitution of the thiazolopyrimidine moiety with methyl group at the 5-position (**11a**) improved activity to some extent. However, substitution with p-chlorophenyl group (**11c**) resulted in broad spectrum of activity against Gram-positive and negative bacteria as well as fungi. The pyridine-2-ones (**8a-c**) had no biological activity. In light of the anti-HIV results of compounds **9a-d**, compound **9a** (R = n-butyl) was confirmed as being moderately active. The activity decreased when R = benzyl (**9b**), phenyl or p-chlorophenyl (**9c**). In conclusion, compounds **5c** and **9a** might be potential candidates for further derivatization as a possible anti-AIDS treatment.

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