Synthesis and anti-microbial activity of (Z)-4-(4-substituted-thiazol-2-yl)-1-(2-oxoindolin-3-ylidene) semicarbazide and its derivatives†

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3-[(2S)-3-Acetyl-5-amino-N-(4-substituted-thiazol-2-yl)2,3-dihydro-1,3,4-oxadiazol-2-yl]-indolin-2-yl acetates, 1-[(N-[2R]-2-(-2-oxoindolin-3-ylidene)-4-oxo-1,3-thiazol-3-yl]-3-(4-substituted-thiazol-2-yl)ureas, N-(4-substituted-thiazol-2-yl)-[1,3,4]oxadiazino[6,5]indol-3-amines and (Z)-4-(4-substituted-thiazol-2-yl)-1-(2-oxo-1-(substituted) methyl)indolin-3-ylidene)semicarbazides have been prepared by making use of (Z)-4-(4-substituted-thiazol-2-yl)-1-(2-oxoindolin-3-ylidene) semicarbazides obtained by the reaction of (4-substituted thiazole-2-yl) semicarbazides and isatin. The structures of the newly synthesized compounds have been confirmed on the basis of analytical, IR, 1H NMR and mass spectral data. The newly synthesized compounds have been screened for their antimicrobial activities. The antimicrobial activities of these newly synthesized compounds have also been reported in this paper.

Keywords: Thiazole, isatin, Schiff base, anti-microbial activity

Heterocycles bearing nitrogen, sulphur and oxygen atoms in their structure constitute the core structure of a number of biologically interesting compounds. Heterocycles containing thiazole rings are associated with a wide range of biological properties such as antiprotozoal, anticonvulsants, a depressant effect on the central nervous system, anti-helminthic, antidiabetics, as inhibitors of dihydrofolate, as inflammation inhibitors, antitumor, herbicides, antimicrobial, antiviral and antianaphylactic activities due to toxophoric –N=C-S- group. Literature survey revealed that a number of isatin derivatives possess various biological properties such as antimicrobial, CNS depressants, anticonvulsants, antipoxvirus, antifertility and antiviral activities. Many oxadiazoles, thiazolodines, oxadiazino and Mannich base derivatives reported in the literature possess good biological activities. In view of these findings and in continuation of our research work on thiazole-2-semicarbazides having various biological activities, we hereby report for the first time the synthesis of some (Z)-4-(4-substituted-thiazol-2-yl)-1-(2-oxoindolin-3-ylidene) semicarbazides and their derivatives viz 3-[(2S)-3-acetyl-5-amino-N-(4-substituted-thiazol-2-yl)2,3-dihydro-1,3,4-oxadiazol-2-yl]-indolin-2-yl acetates 6a,b, 1-[(N-[2R]-2-(-2-oxoindolin-3-ylidene)-4-oxo-1,3-thiazol-3-yl]-3-(4-substituted-thiazol-2-yl)ureas 7a,b, N-(4-substituted-thiazol-2-yl)-[1,3,4]oxadiazino[6,5]indol-3-amines 8a,b and (Z)-1-(1-substituted aminomethyl)-2-oxoindolin-3-ylidene)-4-(4-substituted-thiazol-2-yl)semicarbazides 9a-f by making use of (4-substituted-thiazole-2-yl) semicarbazides 3a,b and isatin 4 as starting materials (Scheme I).

In compounds 6a,b and 8a,b, 4-substituted-thiazole and (2,3-dihydro-1,3,4-oxadiazol-2-yl)-indolin systems and 1,3,4-oxadiazino[6,5]indolo systems are linked together at their 2 and 3 positions respectively via amino bridging. In compounds 7a,b 4-substituted-thiazole and 1-N-[2R]-2-(-2-oxoindolin-3-ylidene)-4-oxo-1,3-thiazolidin] systems are linked to each other at their 2 and 3 positions respectively to two nitrogen atoms of urea moiety. The antimicrobial activities of these newly synthesized compounds have also been reported in this paper.

Experimental Section
The starting materials 4-substituted-2-amino thiazoles 1a,b were prepared according to the reported method. The compounds 2a and 3a were prepared according to the method reported earlier. Compound 3b was also prepared according to the method described by this group for 3a using 1b via intermediacy of ethyl 4-methylthiazole-2-carbmate 2b.
Melting points were determined in open capillary tubes and are uncorrected. IR spectra were recorded (cm⁻¹) in KBr on Perkin-Elmer FT-IR (spectrum 1000) spectrophotometer, ¹H NMR spectra on a Bruker AMX (400 MHz) spectrometer using DMSO-d₆ as solvent using TMS as an internal standard (chemical shifts in δ, ppm) and mass spectra on a mass spectrometer, Joel SX-102 (FAB) instrument.

**Scheme I**

**Synthesis of ethyl 4-methylthiazole-2-carbamate, 2b**

Compound 1b (0.001 mole) was dissolved in minimum amount of pyridine and cooled to 0°C under anhydrous conditions. Ethyl chloroformate (0.001 mole) was added to it dropwise at 0-2°C while stirring under anhydrous conditions. The mixture was stirred at this temperature for 1 hr, further 0.5 hr at RT and...
then heated for 12 hr at reflux on a water-bath. The mixture was then decomposed by pouring into iced-cold water and pyridine removed by steam distillation. The resulting solid was dried and purified by recrystallization from absolute alcohol to get 2b, brown crystals, 75%; m.p. 145°C; IR: 693 (C=S-C), 1597 (C=N), 1723 (C=O), 3187 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{18}\)H\(_{13}\)N\(_2\)O\(_2\): C, 59.50; H, 3.58; N, 19.28. Found: C, 59.06; H, 3.80; N, 15.66. Found: C, 58.98; H, 3.64; N, 15.55%.

**Synthesis of 4-methylthiazole-2-semicarbazide, 3b**

A suspension of 2b (0.001 mole) in ethanol (10 mL) was refluxed with hydrazine hydrate (0.0045 mole, 99%) on a water-bath for 9 hr and then cooled to RT. The resulting white solid separated was filtered, dried and purified by recrystallization from ethanol to offer white crystalline compound 3b, 89%, m.p. 215°C; IR: 700 (C=S-C), 1542 (C=N), 1643 (C=O), 3225, 3278, 3412 cm\(^{-1}\) (NH/NH). Anal. Calcd for C\(_{14}\)H\(_8\)N\(_2\)O\(_2\): C, 47.78; H, 3.38; N, 18.48%. Found: C, 47.09; H, 3.64; N, 18.00%.

**Synthesis of (Z)-4-[(4-substituted-thiazol-2-yl)-1-(2-oxoindolin-3-ylidene)-2-oxoindolin-3-ylidene]-semicarbazides, 5a,b**

Equimolar mixture of (4-substituted-thiazol-2-yl) semicarbazides 3a, 18 (0.001 mole) and isatin 4 (0.001 mole) and a catalytic amount of glacial acetic acid (1-2 drops) in ethanol (20 mL) were refluxed for 7-8 hr on water-bath. The solid separated was filtered, washed with a little alcohol, dried and purified by recrystallization from 1,4-dioxane to get 5a, 17.

**Compound 5a:** yellow crystals, 75%, m.p. 285°C; \(^1\)H NMR: \(\delta\) 6.80 (s, 1H, CH), 7.18-7.52 (m, 9H, Ar-H), 9.54 (s, 1H, NH), 10.21 (s, 1H, NH), 10.80 (s, 1H, NH); MS: \(m/z\) (%) 374 (M\(^+\)), 367, 366, 217 (100), 178. Found: C, 54.68; H, 3.24; N, 18.06%.

**Compound 5b:** yellow needles, 72%, m.p. 260°C; IR: 694 (C=S-C), 1560, 1580 (C=N), 1684, 1719 (C=O), 3188, 3361, 3375 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{20}\)H\(_{13}\)N\(_2\)O\(_2\): C, 59.50; H, 3.58; N, 19.28. Found: C, 59.32; H, 3.57; N, 19.14%.

**Synthesis of 3-[{(2S)-3-acetyl-5-amino-N-(4-substituted-thiazol-2-yl) 2, 3-dihydro-1, 3, 4-oxadiazol-2-yl]}indolin-2-yl acetates, 6a,b**

A mixture of the 5a, b (0.001 mole) and acetic anhydride (10 mL) was refluxed for 3 hr on oil-bath at 145°C. The reaction-mixture was cooled to RT, poured into ice-cold water and the solid separated was filtered, washed with water, dried and purified by recrystallization from ethanol to yield 6a, b.

**Compound 6a:** brown needles, 70%, m.p. 214°C; \(^1\)H NMR: \(\delta\) 2.19 (s, 3H, CH\(_3\)), 2.30 (s, 3H, CH\(_3\)), 7.01 (s, 1H, CH), 7.10-7.62 (m, 9H, Ar-H), 10.35(s, 1H, NH); MS: \(m/z\) (%) 447 (M\(^+\)), 404 (15), 361 (38), 333 (15), 217 (100), 175 (5); IR: 704 (C=S-C), 1155 (C-O-C), 1539, 1601, 1610 (C=N), 1696, 1728 (C=O), 3314 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{25}\)H\(_{17}\)N\(_3\)O\(_3\): C, 59.06; H, 3.80; N, 15.66. Found: C, 58.98; H, 3.64; N, 15.55%.

**Compound 6b:** light brown needles, 72%, m.p. 239°C; IR: 701 (C=S-C), 1159 (C-O-C), 1542, 1583, 1610 (C=N), 1685, 1719 (C=O), 3230 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{25}\)H\(_{17}\)N\(_3\)O\(_3\): C, 52.99; H, 3.90; N, 18.18. Found: C, 52.85; H, 3.74; N, 18.00%.

**Synthesis of 1-[N-[(2R)-2-(-2-oxoindolin-3-ylidene)-4-oxo-1,3-thiazolidin-3-yl]-3-(4-substituted-thiazol-2yl)ureas, 7a,b**

Compound 5a, b (0.001 mole) was refluxed in dimethylformamide (30 mL) containing a pinch of anhydrous zinc chloride and thioglycolic acid (0.001 mole) for 8 hr. The reaction-mixture was cooled to RT and poured into ice-cold water. The separated solid was filtered, washed with a little dimethylformamide, then with water, dried and purified by recrystallization from isopropyl alcohol to get 7a, b.

**Compound 7a:** brown needles, 59%, m.p. 197°C; \(^1\)H NMR: \(\delta\) 3.32 (s, 2H, CH\(_2\)), 6.88 (s, 1H, CH), 6.98-7.54 (m, 9H, Ar-H), 9.83 (s, 1H, NH), 10.24 (s, 1H, NH), 10.80 (s, 1H, NH); MS: \(m/z\) (%) 437 (M\(^+\)), 436 (35), 391 (15), 365 (19), 245 (27), 219 (100), 203 (57); IR: 723, 748 (C=S-C), 1618 (C=N), 1649, 1678, 1720 (C=O), 3135, 3246, 3298 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{26}\)H\(_{13}\)N\(_2\)O\(_2\): C, 54.92; H, 3.43; N, 17.06. Found: C, 54.79; H, 3.31; N, 16.86%.

**Compound 7b:** brown needles, 67%, m.p. 218°C; IR: 727, 754 (C=S-C), 1614 (C=N), 1655, 1685, 1747 (C=O). Anal. Calcd for C\(_{26}\)H\(_{13}\)N\(_2\)O\(_2\): C, 51.83; H, 3.65; N, 23.26. Found: C, 51.68; H, 3.45; N, 23.15%.

**Synthesis of N-(4-substituted-thiazol-2-yl)-[1, 3, 4] oxadiazino[6,5]indol-3-amines, 8a,b**

Compound 5a, b (0.001 mole) was added slowly to concentrated sulphuric acid (AR grade, 0.015 mole)
in the cold, while stirring and the stirring was continued for 0.5 hr more. The resulting mass was allowed to attain RT and poured into ice-cold water. After neutralization with liquid ammonia, the product(s) \( 8a, b \) separated was collected by filtration, washed with water, dried and purified by recrystallization from 1,4-dioxane.

**Compound 8a:** yellow crystals, 74%, m.p. 231°C; \(^1\)H NMR: \( \delta \) 7.11-7.76 (m, 10H, Ar-H + CH), 12.72 (s, 1H, NH); MS: \( m/z \) (%) 345 (M\(^+\)), 100, 303 (42), 174 (76), 116 (40); IR: 737 (C=S-C), 1150 (C=O-C), 1574, 1616, 1629, 1654 (C=N), 3276 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{16}\)H\(_{10}\)N\(_2\)O\(_2\): C, 55.01; H, 3.14; N, 11.14%.

**Compound 8b:** yellow needles, 75%, m.p. 261°C; IR: 737 (C=S-C), 1150 (C=O-C), 1576, 1616, 1631, 1658 (C=N), 3282 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{18}\)H\(_{11}\)N\(_2\)O\(_2\): C, 55.10; H, 3.21; N, 20.29. Found: C, 55.01; H, 3.14; N, 20.08%.

**Results and Discussion**

4-Phenylthiazole-2-semicolonbazide 3a was prepared according to the procedure reported by us\(^\text{13}\). 4-Methylthiazole-2-semicolonbazide 3b was prepared starting from 4-methyl-2-aminothiazole\(^\text{43}\) via the intermediacy of ethyl 4-methylthiazole-2-carbamate 2b, by the procedure described for 3a elsewhere\(^\text{13}\). The IR spectrum of compound 2b displayed absorption peaks at 693, 1597, 1723 and 3187 cm\(^{-1}\) due to C=S-C, C=N, C=O and NH functions respectively, which is in conformity with its structure 2b.

**Compound 9c:** greenish yellow needles, 68%, m.p. 256°C; \(^1\)H NMR: \( \delta \) 2.22-2.51 (m, 6H, -CH\(_2\)-CH\(_2\)-CH\(_2\)-), 2.84 (t, 4H, -CH\(_2\)-N-CH\(_2\)-), 3.40 (s, 2H, -N-CH\(_2\)-N-), 7.51 (s, 1H, CH), 7.69-8.20 (m, 9H, Ar-H), 10.12 (s, 1H, NH), 10.80 (s, 1H, NH); IR: 749 (C=S-C), 1578, 1614 (C=N), 1661, 1686 (C=O), 3110, 3355 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{24}\)H\(_{22}\)N\(_2\)O\(_2\): C, 62.61; H, 5.22; N, 18.26. Found: C, 62.46; H, 5.09; N, 18.17%.

**Compound 9d:** brown amorphous needles, 72%, m.p. 188°C; IR: 721 (C=S-C), 1580, 1601 (C=NH), 1682, 1737 (C=O), 3320, 3375 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{19}\)H\(_{18}\)N\(_2\)O\(_2\): C, 53.63; H, 5.03; N, 23.46. Found: C, 53.53; H, 4.86; N, 23.31%.

**Compound 9e:** brown needles, 69%, m.p. 207°C; IR: 721 (C=S-C), 1598, 1603 (C=NH), 1664, 1703 (C=O), 3335, 3360 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{19}\)H\(_{22}\)N\(_2\)O\(_2\): C, 55.96; H, 5.70; N, 21.76. Found: C, 55.77; H, 5.64; N, 21.68%.

**Compound 9f:** light yellow needles, 75%, m.p. 284°C; IR: 701 (C=S-C), 1598, 1620 (C=NH), 1678, 1703 (C=O), 3245, 3305 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{19}\)H\(_{22}\)N\(_2\)O\(_2\): C, 57.29; H, 5.53; N, 21.10. Found: C, 57.12; H, 5.43; N, 20.95%.

**Synthesis of (Z)-1-(1-substituted aminomethyl)-2-oxindolin-3-ylidene)-4-(4-substituted-thiazol-2-yl)semicolonbazides, 9a-f**

Compound 5a,b (0.001 mole) was suspended in a minimum quantity of dimethyl formamide (10 mL). To these solutions, formaldehyde (0.001 mole) and various secondary amines (0.001 mole) were added with vigorous stirring. The reaction-mixture was heated on water-bath for 20 min, and left overnight. The products \( 9a-f \) separated were collected by filtration, washed with little dimethylformamide and purified by recrystallization from 1,4-dioxane.

**Compounds 9a:** brown crystals, 59%, m.p. 252°C; \(^1\)H NMR: \( \delta \) 2.30 (s, 6H, -N-(CH\(_2\)_2), 3.61 (s, 2H, -N-CH\(_2\)-N-), 7.14 (s, 1H, CH), 7.21-7.68 (m, 9H, Ar-H), 10.14 (s, 1H, NH), 10.30 (s, 1H, NH); MS: \( m/z \) (%) 420 (M\(^+\)), 362 (33), 319 (5), 203 (68); IR: 723 (C=S-C), 1584, 1620 (C=NH), 1682, 1722 (C=O), 3130, 3244 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{21}\)H\(_{20}\)N\(_2\)O\(_2\): C, 60.00; H, 4.76; N, 20.00. Found: C, 59.78; H, 4.71; N, 19.81%.

**Compounds 9b:** brown crystals, 69%, m.p. 174°C; \(^1\)H NMR: \( \delta \) 2.14 (t, 6H, 2CH\(_3\)), 3.45 (q, 4H, 2CH\(_2\)), 3.81 (s, 2H, -N-CH\(_2\)-N-), 6.92-7.68 (m, 10H, Ar-H and CH), 10.34 (s, 1H, NH), 10.80 (s, 1H, NH); IR: 723 (C=S-C), 1582, 1615 (C=NH), 1682, 1722 (C=O), 3232, 3382 cm\(^{-1}\) (NH). Anal. Calcd for C\(_{22}\)H\(_{20}\)N\(_2\)O\(_2\): C, 61.61; H, 5.36; N, 18.75. Found: C, 61.50; H, 5.21; N, 18.62%.
C=N/C=N, C=O/C=O and NH/NH/NH functions respectively. Its $^1$H NMR spectrum displayed three singlets at δ 9.54, 10.21 and 10.80 due to two NH protons of urea moiety and a proton of NHCO functions respectively. Nine aromatic protons and a proton of thiazole at 5-position have resonated as multiplet and singlet at δ 7.18-7.52 and 6.80 respectively. Mass spectral fragmentation pattern of compound 5a displayed molecular ion peak $M^+$ at 363 (45%) which is equivalent to its molecular weight. Further, it showed M+1 peak at 364 (100%) which is also a base peak. Fragment ions observed at $m/z$ 273 (6%), 232 (30%), 203 (27%), 161 (22%) and 136 (83%) are due to the sequential expulsion of C$_6$H$_4$N radical, C$_2$HO radical, simultaneous loss of hydrogen radical and nitrogen molecule followed by the loss of NCO radical, respectively from the molecular ion. The fragment ion recorded at $m/z$ 232, due to simultaneous expulsion of N$_2$, NCO and CN species gave a fragment ion observed at $m/z$ 136 (83%) (Scheme II). All these data are in conformity with the structure 5a.

Compounds 5a,b when reacted with acetic anhydride under reflux conditions on oil-bath at
145°C for 3 hr underwent cyclization followed by simultaneous acetylation of NH function of oxadiazoline moiety and enolised form of 2-oxo-indoline moiety of the resultant product of cyclization to afford 3-[(2S)-3-acetyl-5-amino-N-(4-substituted-thiazol-2-yl)-2,3-dihydro-1,3,4-oxadiazol-2-yl]-indolin-2-yl acetates 6a,b in good yield. Compound 6a in its IR spectrum showed absorption bands at 704, 1155, 1539, 1601, 1696, 1728 and 3314 cm\(^{-1}\) due to (C-S-C), (C-O-C), C=N/C=N/C=N, C=O/C=O and NH functions respectively. Four singlets and a multiplet, observed at \(\delta\) 2.19, 2.30, 7.01, 10.35 and 7.10-7.62 in the \(^1\)H NMR spectrum of compound 6a were due to the methyl protons of N-acetyl group on oxadiazoline moiety, methyl protons of carboxyl methyl function at 2-position of indoline moiety, a proton at 5-position of thiazole moiety, a proton of bridged NH and nine aromatic protons respectively.

Mass spectrum of compound 6a exhibited molecular ion peak M\(^+\) at 447 (51%) which corresponds to its molecular weight. Due to the sequential loss of acetyl radical, CO molecule, simultaneous expulsion of \(\text{C}_6\text{H}_4\text{N}\) radical and CN radical followed by the loss of NCO radical respectively from the molecular ion of compound 6a gave fragment ions recorded at \(m/z\) 404 (15%), 361 (38%), 333 (15%), 217 (100%, base peak) and 175 (5%) respectively (Scheme III). All these data prove the formation of compound 6a from compound 5a.

Compounds 5a,b when allowed to react with thioglycolic acid in dimethylformamide in presence of catalytic amount of anhydrous zinc chloride furnished 1-\{N-[(2R)-2-(2-oxoindolin-3-ylidene)-4-oxo-1,3-thiazolidin-3-yl]-4-(substituted-thiazol-2-yl)ureas 7a,b in good yield. The IR (\(\nu\), in cm\(^{-1}\)) spectrum of 7a

\[\text{Scheme III}\]
exhibited absorption bands at 723, 748, 1618, 1649, 1678, 1720, 3135, 3246 and 3298 cm\(^{-1}\) due to C=S-C/C=S-C, C=N, C=O/C=O/ C=O and NH/NH/NH functions respectively. In the \(^1\)H NMR spectrum of 7\(a\) (in \(\delta,\) ppm) two methylene protons of thioazolidinone moiety, nine aromatic protons, a proton at 5-position of thiazole moiety, proton on indole NH and two NH protons of urea function have resonated as singlet at \(\delta\) 3.32, multiplet in the region 6.98-7.54, singlets at \(\delta\) 6.88, 9.83, 10.24 and 10.80 respectively. Mass spectrum 7\(a\) displayed molecular ion peak M\(^+\) at 437 (55%) which is equivalent to its molecular weight. Further, fragment ions recorded at \(m/z\) 436 (35%), 391 (15%), 363 (19%), 245 (27%), 219 (100%, base peak) and 203 (57%) are due to the sequential expulsion of H radical, HCS radical, CO molecule, simultaneous expulsion of C\(_6\)H\(_4\)N radical and CO molecule, followed by sequential expulsion of CN and NH\(_2\) radicals respectively from the molecular ion (Scheme IV). These data clearly prove the formation of compound 7\(a\) from 5\(a\).
Compounds 5a,b on treatment with concentrated sulphuric acid at RT followed by treatment with ammonia gave cyclized product N-(4-substituted-thiazol-2-yl)-(1,3,4]oxadiazino[6,5]indol-3-amine 8a,b in good yield. Compound 8a in its IR spectrum showed absorption bands at 737, 1150, 1574, 1616, 1629, 1654 and 3276 cm$^{-1}$ due to the C-S-C, C-O-C, C=N/C=N/C=N/C=N and NH functions respectively. A singlet and a multiplet observed at $\delta$ 12.72 and 7.11-7.76 in $^1$H NMR spectra of compound 8a are due to one proton of NH function and nine aromatic protons and a proton of thiazole moiety at its 5-position respectively. Mass spectrum of compound 8a exhibited molecular ion peak M$^+$ at 345 (100%) which is equivalent to its molecular weight and is also a base peak. Further, fragment ions recorded at $m/z$ 303 (42%), 174 (76%) and 116 (40%) respectively. These spectral data clearly prove the formation of compound 8a from 5a.

Compounds 5a,b, when allowed to react with formaldehyde and secondary amines in minimum amount dimethylformamide offered (Z)-1-(1-substituted-aminomethyl)-2-oxindolin-3-ylidene-4-(4-substituted-thiazol-2-yl)semicarbazides 9a-f in good yield. Compound 9a in its IR spectrum showed absorption bands at 723, 1584, 1620, 1682, 1722, 3130 and 3244 cm$^{-1}$ due to C-S-C, C=N/C=N, C=O/C=O and NH functions respectively. In the $^1$H NMR of spectrum (in $\delta$) of compound 9a exhibited a singlet at $\delta$ 2.30 accounting for six methyl protons of N-dimethyl function, a singlet at $\delta$ 3.61 for bridged two methylene protons attached to indole nitrogen and a multiplet appeared at $\delta$ 7.21-7.68 for nine aromatic protons. Three singlets appeared at $\delta$ 7.14, 10.14 and 10.30 were due to a proton at 5-position of thiazole and two protons of urea moiety of semicarbazone function respectively. Mass spectrum 9a displayed molecular ion peak M$^+$ at 420 (100%, base peak) which is equivalent to its molecular weight and is also a base peak. Further, fragment ions recorded at $m/z$ 362 (33%), 319 (5%) and 203 (68%) are due to sequential expulsion of C$_3$H$_8$N radical, simultaneous expulsion of hydrogen and NCO radicals followed by simultaneous expulsion of CN and C$_6$H$_4$N radicals respectively from the molecular ion (Scheme VI). These data clearly prove the formation of compound 9a from 5a.

**Antimicrobial activity**

The in vitro biological screening of the synthesized compounds was undertaken against the bacterial species namely, *Staphylococcus aureus*, *Escherichia coli* and *Bacillus subtilis* and fungi species namely,
Aspergillus niger and Candida albicans by cup-plate method\textsuperscript{13,14} using nutrient agar medium. The holes of 6 mm diameter were punched carefully using a sterile cork borer and these were filled with test solutions (1000 µg/mL in DMF) and DMF was used as control. The plates were incubated at 37°C for 24 hr and 72 hr in case of antibacterial and antifungal activity, respectively. The diameter of the zone of inhibition for all the test compounds was measured and the results were compared with that of standard drug Gentamycin for antibacterial activity and Nystatin for antifungal activity (Table I).
The results showed that the compounds 7a, 7b, 8b, 9a and 9c showed good activity, compounds 5b and 6a exhibited moderate activity against S. aureus when compared with standard drug Gentamycin. Compounds 6a, 7a, 8b and 9c showed good activity, compounds 7b, 9a and 9e showed moderate activity when compared to Gentamycin against E. coli. Compounds 7a, 7b, 8b, 9a and 9c showed good activity, compounds 6a, 8a and 9d exhibited moderate activity against B. subtilis when compared to Gentamycin. Compounds 9c and 9a showed good activity, compounds 5b and 7a exhibited moderate activity when compared to Nystatin against A. Niger. Compounds 5b, 7a, 9a and 9c showed good activity, compounds 8b and 9d exhibited moderate activity when compared to Nystatin against C. albicans. Rest of the compounds showed lower activity against all the microorganisms tested when compared to that of standard drugs at the same concentration as that of test compounds.

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References