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Conventional and Microwave Assisted Synthesis of Some Novel 2-(Substituted phenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazoles of Biological Interest

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In present investigation, a straightforward approach to design and synthesize 2-(substituted phenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazoles (**5a-j**) using two methods *viz*. traditional heating and microwave irradiation and characterized with elemental, FT-IR, ^{1}H NMR, ^{13}C NMR, mass spectrophotometric techniques. The microwave irradiation method offers excellent yields, lesser reaction time and eco-environmental friendly reactions as compared to conventional method. The synthesized compounds were evaluated for their antimicrobial, anticancer and free radical scavenging activity. The findings on the compounds' *in vitro* antibacterial activity against Gram-positive and Gram-negative pathogens showed that compounds **5b**, **5c** and **5g** showed potential antibacterial and fungicide efficacy. Using Dalton's Lymphoma Ascites (DLA) cell lines, the synthesized compounds were further examined for its *in vitro* anticancer efficacy. Among the synthesized compounds, **5b**, **5f**, **5g** and **5j** exhibited excellent anticancer activity with CTC_{50} value of 31.25 µg/mL, 51.61 µg/mL, 44.21 µg/mL and 31.25 µg/mL, respectively. The synthesized compounds have also shown a marked free radical scavenging capacity in all the concentrations but specifically compounds **5a**, **5d**, **5e**, **5f**, **5i** and **5j** have shown good antioxidant potential with an IC_{50} value of 25.18 µmol/L, 44.22 µmol/L 35.61 µmol/L, 28.09 µmol/L, 44.47 µmol/L and 39.46 µmol/L, respectively.

Keywords: Imidazo-thiazole, Microwave assisted synthesis, Antibacterial, Anticancer, Free radical scavenging.

INTRODUCTION

Heterocycles are highly important pharmacophores in the fields related to pharmaceutical and drug discovery studies [1-4]. According to the World Health Organization (WHO), heterocycles make up around 90-95% of the drugs available in the market. Heterocycles, which include thiazoles and imidazole, are the vital broad-spectrum medicines owing to their five-membered ring structure [5-8]. Both molecules possess distinctive physico-chemical features, including low basicity, a significant dipole moment that facilitates π-π stacking interactions and strong dual hydrogen-bonding capability, which can be significant in drug-target interactions [9-11].

Imidazole containing molecules such as metronidazole is used as antimicrobial drug [12], cimetidine is used as peptic ulcer [13], omeprazole as anti-inflammatory and methotrexate as anticancer agents [14]. Similarly, thiazole containing molecules such as sulfathiazole are used as an antimicrobial drug [15], ritonavir as antiretroviral drug [16], abafungin as antifungal drug [17] and tiazofurin as antineoplastic drug [18]. Thiazole is also present in penicillin which is the first one of the broad-spectrum antibiotic [19].

The current work aims to design, synthesize and evaluate the therapeutic effects as well as the current strategies used in the synthesis of dihybrid derivatives of imidazole and thiazole employing various angles. The presence of two different biologically active heterocycles in such scaffolds proved to be valuable and beneficial as it improved their therapeutic properties [20], hence in this work, we have synthesized imidazo-thiazole hybrid derivatives **5a-j** and characterized. All the synthesized compounds were also evaluated for biological activities *viz*. *in vitro* antimicrobial activities by agar well diffusion method against the selected strains, antioxidant activities by using the DPPH assay method and the cytotoxicity activity against Dalton's Lymphoma Ascites (DLA) cell lines.

EXPERIMENTAL

All chemicals and solvents were purchased commercially and used as such. The melting points were measured using open capillaries and are uncorrected. The FT-IR spectral analysis was conducted using Perkin-Elmer infrared-283 spectrophotometer with KBr pellets techniques. Using aluminum plates 60 F_{254} with silica gel coating of 0.25 mm thickness, the progress of the synthesized compounds were monitored. The ^{1}H NMR and ^{13}C NMR spectra were recorded using a Bruker DRX-400 spectrophotometer operating at 400 MHz and 75 MHz, respectively using TMS as an internal standard and the solvents were $CDCl_3$ and DMSO-d_6. Instrument Elemental Vario EL III, Carlo Erba 1108 was used for the elemental analysis and mass spectrum was acquired using LC-MS instrument (Shimadzu-2010).

Synthesis of target molecule 2-(substituted phenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazole derivatives (**5a-j**) using conventional as well as microwave methods.

Conventional synthesis of Schiff bases (3a-j): In a 500 mL flat bottom flask, a mixture of thiazole-2-amine (**1**, 0.01 M) and substituted aromatic aldehyde (**2**, 0.01 M) in 30 mL of glacial acetic acid was refluxed for 7 h. After the completion of the reaction (monitored by TLC) and cooled to room temperature, the obatined Schiff base was used for the synthesis of imidazo-thiazole hybrid derivatives.

Conventional synthesis of 2-(substituted phenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazoles (5a-j): In a 500 mL round-bottom flask, a mixture of benzil (**4**, 0.01 M), ammonium acetate (0.1 M) and Schiff base (**3**, 0.01 M) was refluxed for 10-12 h. As the reaction has completed (monitored by TLC), the excess ammonium acetate and acetic acid (byproduct) were eleminated by filtration. The solid product was poured into 200 mL of ice-cold water and cooled to room temperature, filtrated then washed with 30 mL of benzene to remove traces of any unreacted benzil. The product was finally recrystallized by chloroform to obtain the substituted analogues of imidazo-thiazole **(5a-j)**.

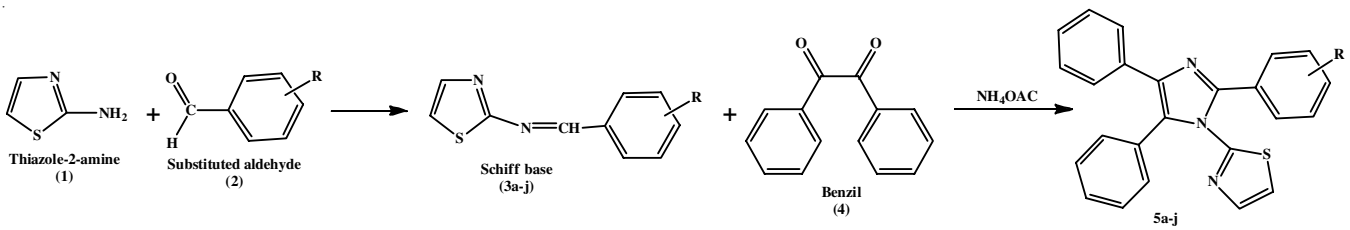
Microwave synthesis of Schiff bases (3a-j): A homogenous mixture was formed by triturating a mixture of thiazole-2-amine (**1**, 0.01 M) and substituted aromatic aldehyde (**2**, 0.01 M) in a dry and clean mortar. The mixture was then transferred to different 100 mL beaker containing 5 g of activated silica gel. The microwave oven was used to irradiate the beaker holding various reaction mixtures for approximately 8 min at 1000 W. After every 60 s, there was an intermittent cooling period following the microwave irradiation. The reaction mixture was thoroughly mixed and periodically cooled. As the reaction has completed (monitored by TLC), the obtained solid product was directly utilized for the synthesis of analogues of imidazo-thiazole.

Microwave synthesis of 2-(substituted phenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazoles (1b-10b): In a dry, clean mortar, a mixture of benzil (**4**, 0.01 M), ammonium acetate (0.1 M) and Schiff base (**3**, 0.01 M) was triturated to obtain a uniform slurry. The uniform slurry (reaction mixture) was put into a 100 mL beaker was exposed to microwave radiation for 12-16 min at 1000 W. After every 60 s, there was an intermittent cooling period following microwave irradiation. After the completion of the reaction, The product was transferred into 250 mL ice-cold water container in order to remove excess ammonium acetate and acetic acid (byproduct) (**Scheme-I**). The resulting precipitate was collected by filtration, washed thoroughly with chloroform followed by 20 mL of benzene to eliminate the unreacted benzil and finally recrystallized by chloroform.

2-(2,6-Dichlorophenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazole (5a): Yellow crystals; m.p.: 226 ºC; IR (KBr, ν_{max}, cm^{-1}): 3107 (Ar C-H), 1640 (C=C), 1537 (C=N), 1275 (C-N), 1165 (C-S), 765 (C-Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 6.90-7.80 (14H, m, Ar-H), 8.20 (1H, d, CH); ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm: 118.1, 121.9, 127.9 (6C), 128.6 (2C), 129.1 (4C), 129.8, 131.0 (2C), 131.9, 134.1 (2C), 137.0, 143.1, 144.1, 152.9; EIMS (*m/z*): $[M]^+$ 449.12, $[M+2]^+$ 450.68; Fragments: 413.08, 379.11, 371.01, 364.05, 337.04, 303.08, 294.97, 288.02, 211.99, 180.09, 159.98, 126.02, 100.01, 78.05; Elemental analysis of calcd. (found) % of $C_{24}H_{15}N_3SCl_2$: C, 64.29 (64.31); H, 3.37 (3.44); Cl, 15.81 (15.68); N, 9.37 (9.29); S, 7.15 (7.10).

2-(3-Methoxyphenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazole (5b): White crystals; m.p. 172 ºC; IR (KBr, ν_{max}, cm^{-1}): 3058 (Ar C-H), 1645 (C=C), 1555 (C=N), 1251 (C-N), 1209 (C-O-C), 1160 (C-S); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 3.02 (S, 3H, OCH_3), 6.85-7.72 (15H, m, Ar-H), 8.12 (1H, d, CH); ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm: 56.0, 112.1, 115.0, 118.9, 120.0, 121.8, 127.6 (4C), 128.9 (2C), 129.2 (4C), 129.8, 130.1, 131.8, 133.4 (2C), 143.0, 144.0, 152.8, 161.5; EIMS (*m/z*): $[M]^+$ 409.96; Fragments: 379.11, 333.09, 326.14, 303.08, 257.06, 250.11, 180.09, 174.08, 122.07, 100.01, 78.05; Elemental analysis of calcd. (found) % of $C_{25}H_{19}N_3OS$: C, 73.32 (73.25); H, 4.68 (4.61); N, 10.26 (10.30); O, 3.91 (3.95); S, 7.83 (7.86).

2-(4-Dimethylaminophenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazole (5c): Yellow amorphous solid; m.p. 220 ºC; IR (KBr, ν_{max}, cm^{-1}): 3022 (Ar C-H), 1670 (C=C), 1540 (C=N), 1229 (C-N), 1172 (C-S); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.97 (s, 6H, $N(CH_3)_2$), 6.83-7.67 (15H, m, Ar-H), 8.15 (1H, d, CH); ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm: 40.8 (2C), 115.1



Scheme-I: Synthetic route of 2-(substituted phenyl)-4,5-diphenyl-1-thiazol-2-yl)-1*H*-imidazoles (**5a-j**)

(2C), 119.2, 120.8, 121.9, 127.0 (4C), 128.9 (2C), 129.3 (2C), 129.8 (4C), 130.1, 133.3 (2C), 143.1, 143.7, 150.0, 152.7; EIMS (*m/z*): $[M]^+$ 423.15; Fragments: 379.11, 346.13, 339.17, 303.08, 270.09, 263.14, 187.11, 180.09, 135.10, 100.01, 78.05; Elemental analysis of calcd. (found) % of $C_{26}H_{22}N_4S$: C, 73.90 (73.99); H, 5.25 (5.10); N, 13.26 (13.34); S, 7.59 (7.49).

2-(4-Chlorophenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazole (5d): Yellow crystals; m.p. 189 ºC; IR (KBr, ν_{max}, cm^{-1}): 3098 (Ar C-H), 1665 (C=C), 1562 (C=N), 1229 (C-N), 1168 (C-S), 745 (C-Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 6.78-7.59 (15H, m, Ar-H), 8.13 (1H, d, CH); ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm: 119.1, 121.9, 128.0 (4C), 129.1 (3C), 129.4 (2C), 129.6 (4C), 129.5 (2C), 129.9, 133.4 (2C), 134.5, 143.0, 144.1, 153.1; EIMS (*m/z*): $[M]^+$ 414.93, $[M+2]^+$ 416.10; Fragments: 379.11, 337.04, 330.09, 303.08, 261.01, 254.06, 180.09, 178.03, 126.02, 100.01, 78.05; Elemental analysis of calcd. (found) % of $C_{24}H_{16}N_3SCl$: C, 69.64 (69.56); H, 3.90 (3.85); Cl, 8.57 (8.70); N, 10.15 (10.20); S, 7.75 (7.80).

2-(4-Fluorophenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazole (5e): Radish black crystals; m.p. 176 ºC; IR (KBr, ν_{max}, cm^{-1}): 3050 (C-H), 1655 (C=C), 1566 (C=N), 1248 (C-N), 1202 (C-F), 1172 (C-S); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 6.89-7.78 (15H, m, Ar-H), 8.17 (1H, d, CH); ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm: 115.8 (2C), 118.9, 121.9, 126.6, 127.9 (4C), 129.0 (2C), 129.6 (2C), 129.9 (4C), 130.0, 133.5 (2C), 142.6, 143.9, 153.0, 163.1; EIMS (*m/z*): $[M]^+$ 398.11; Fragments: 379.11, 321.07, 314.12, 303.08, 245.04, 238.09, 180.09, 100.01, 110.05, 162.06, 78.05; Elemental analysis of calcd. (found) % of $C_{24}H_{16}N_3SF$: C, 72.52 (72.40); H, 4.06 (4.13); F, 4.78 (4.81); N, 10.57 (10.47); S, 8.07 (8.10).

2-(4-Nitrophenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazole (5f): Cream colour crystals; m.p. 181 ºC; IR (KBr, ν_{max}, cm^{-1}): 3046 (C-H), 1664 (C=C), 1575 (C=N), 1443 (N=O), 1251 (C-N), 1156 (C-S); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 6.91-7.79 (15H, m, Ar-H), 8.23 (1H, d, CH); ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm: 118.9, 122.1 (2C), 122.5, 127.7 (4C), 128.7 (2C), 128.9 (2C), 129.6 (4C), 129.9, 132.9 (2C), 137.0, 143.0, 144.0, 148.8, 152.6; EIMS (*m/z*): $[M]^+$ 424.93; Fragments: 379.11, 348.07, 341.12, 303.08, 272.04, 265.09, 189.05, 180.09, 137.05, 100.01, 78.05; Elemental analysis of calcd. (found) % of $C_{24}H_{16}N_4O_2S$: C, 67.91 (67.85); H, 3.80 (3.72); N, 13.20 (13.30); O, 7.54 (7.45); S, 7.55 (7.51).

2-(4-Methoxyphenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazole (5g): White amorphous solid; m.p. 191 ºC; IR (KBr, ν_{max}, cm^{-1}): 3062 (Ar C-H), 1675 (C=C), 1568 (C=N), 1245 (C-O), 1229 (C-N), 1165 (C-S); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 3.06 (s, 3H, OCH_3), 6.89-7.71 (15H, m, Ar-H), 8.21 (1H, d, CH); ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm: 56.0, 115.1 (2C), 119.1, 122.2, 123.6, 127.6 (4C), 128.6 (2C), 128.9 (2C), 129.2 (4C), 129.6, 132.9 (2C), 142.6, 143.2, 152.6, 161.1; EIMS (*m/z*): $[M]^+$ 410.30; Fragments: 379.11, 333.09, 326.14, 303.08, 257.06, 250.11, 180.09, 174.08, 122.07, 100.01, 78.05; Elemental analysis of calcd. (found) % of $C_{25}H_{19}N_3OS$: C, 73.32 (73.45); H, 4.68 (4.70); N, 10.26 (10.30); O, 3.91 (3.92); S, 7.83 (7.88).

2,4,5-Triphenyl-1-(thiazol-2-yl)-1*H*-imidazole (5h): White crystals; m.p. 142 ºC; IR (KBr, ν_{max}, cm^{-1}): 3026 (C-H), 1650 (C=C), 1548 (C=N), 1252 (C-N), 1148 (C-S); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 6.83-7.80 (16H, m, Ar-H), 8.17 (1H, d, CH); ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm: 118.9, 121.8, 127.6 (6C), 128.6 (3C), 129.5 (6C), 129.9, 131.0, 132.8 (2C), 143.0, 144.0, 152.9; EIMS (*m/z*): $[M]^+$ 380; Fragments: 303.08, 296.13, 227.05, 220.10, 180.09, 144.07, 100.01, 78.05; Elemental analysis of calcd. (found) % of $C_{24}H_{17}N_3S$: C, 75.96 (75.91); H, 4.52 (4.45); N, 11.07 (11.10); S, 8.45 (8.50).

2-(3-Chlorophenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazole (5i): Yellowish crystals; m.p. 197 ºC; IR (KBr, ν_{max}, cm^{-1}): 3102 (C-H), 1652 (C=C), 1567 (C=N), 1240 (C-N), 1167 (C-S), 750 (C-Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 6.89-7.71 (15H, m, Ar-H), 8.16 (1H, d, CH); ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm: 118.3, 121.9, 125.8, 126.9, 127.9 (4C), 129.2 (2C), 129.7, 130.4 (4C), 130.9, 131.4, 132.5, 133.5 (2C), 134.6, 143.0, 144.1, 152.8; EIMS (*m/z*): $[M]^+$ 414.12, $[M+2]^+$ 415.92; Fragments: 379.11, 337.04, 330.09, 303.08, 261.01, 254.06, 180.09, 178.03, 126.02, 100.01, 78.05; Elemental analysis of calcd. (found) % of $C_{24}H_{16}N_3SCl$: C, 69.64 (69.69); H, 3.90 (3.92); Cl, 8.57 (8.50); N, 10.15 (10.01); S, 7.75 (7.68).

2-(3-Nitrophenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazole (5j): Pale yellow crystals; m.p. 185 ºC; IR (KBr, ν_{max}, cm^{-1}): 3081 (C-H), 1648 (C=C), 1562 (C=N), 1463 (N=O), 1252 (C-N), 1172 (C-S); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 6.80-7.72 (15H, m, Ar-H), 8.15 (1H, d, CH); ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm: 118.9, 120.8, 122.2, 122.7, 127.2 (4C), 129.0 (2C), 129.8 (4C), 130.3, 130.8, 131.2 (2C), 132.9 (2C), 142.6, 144.1, 148.8, 153.1; EIMS (*m/z*): $[M]^+$ 425.23; Fragments: 379.11, 348.07, 341.12, 303.08, 272.04, 265.09, 189.05, 180.09, 137.05, 100.01, 78.05; Elemental analysis of calcd. (found) % of $C_{24}H_{16}N_4O_2S$: C, 67.91 (67.86); H, 3.80 (3.74); N, 13.20 (13.25); O, 7.54 (7.49); S, 7.55 (7.51).

Biological studies

Antimicrobial activity: The agar disk-diffusion method was used to evaluate the *in vitro* antibacterial and antifungal activities [21-23]. Following the recommendations of CLSI standards, the culture medium, incubation conditions and interpretation criteria for inhibition zones were observed and analyzed. The experiment was conducted by applying microbial inoculums of around $1\text{-}2 \times 10^8$ CFU/mL to 150 mm solid agar plate. Over the agar solid surface, the antimicrobial agents were added in different concentrations and covered with filter paper with a diameter of approximately 12 mm. The chosen microbe was inoculated into agar plates. Next, filter paper discs with a diameter of roughly 6 mm were put on top of the agar. The synthesized scompound and a selected microorganisms were added topetri dishes and then incubated at 27 ºC. The zone diameter indicating the inhibition of growth of the microbial strain was assessed by the nearest millimeters.

Anticancer activity: As per usual protocol, the anticancer activity of the synthesized compounds was evaluated by calculating the percentage growth inhibition of the DLA cell lines using the Trypan blue dye exclusion test [24-26]. Each synthesized compounds was tested for its anticancer efficacy at doses of 500, 250, 125, 62.5 and 31.25 μg/mL and the standard drug used was vincristine (CTC_{50}, 12.25 μg/mL). Eqn. 1 was used to calculate the percentage growth inhibition.

$$\text{Growth inhibition (\%)} = \frac{\text{Total cell} - \text{Live cell}}{\text{Total cells}} \times 100 \quad (1)$$

Plotting the concentration against the percentage growth inhibition and bisecting the concentration at the 50% growth inhibition obtained the CTC_{50} value.

Antioxidant activity: The efficacy of each synthesized compound to scavenge free radicals was assessed using 0.1 mmol DPPH solution in methanol [27,28]. Solutions were kept in darkness for 30 min to form free radicals. The solution of test compounds and standard were prepared with varying concentrations (10, 20, 30, 40 and 50 µg/mL). Then, 1 mL of each test compound solution was added with the same volume of DPPH solution and then the mixture was mixed vigorously and kept for 30 min in the darkroom. The absorbance of each solution was measured at the wavelength of 517 nm. The same process was carried out in triplicate (n = 3) and the results include a standard deviation for the average of three readings. Ascorbic Acid was used as standard drug and eqn. 2 was used to calculate the percent inhibition.

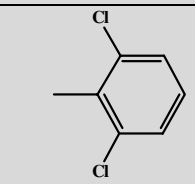
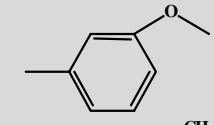
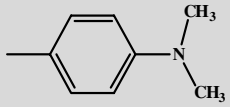
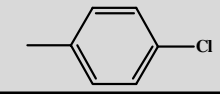
$$\text{Inhibition (\%)} = \frac{A_{control} - A_{sample}}{A_{control}} \times 100 \quad (2)$$

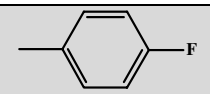
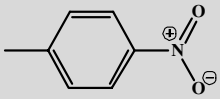
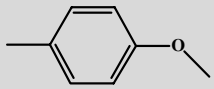
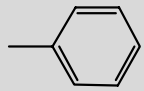
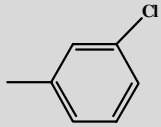
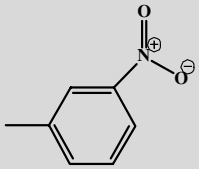
RESULTS AND DISCUSSION

The IR spectra of the newly synthesized compounds **5a-j** showed the presence of characteristic C-H stretching (aromatic) vibrations gave rise to a band at 3107-3026 cm^{-1}. The stretching bands for C-S and C-Cl were observed between 1148-1172 & 745-765 cm^{-1}, respectively. The characteristic absorption bands in the region 1675-1640 cm^{-1} for C=C, 1575-1537 cm^{-1} for aromatic C=N stretching and 1275-1229 cm^{-1} for C-N stretching, respectively. The stretching of C-F bonds was observed at 1202 cm^{-1} in compound **5e**, whereas the for C-Cl bonds were appeared in the 765-745 cm^{-1} in compounds **5a**, **5d** and **5i**. In the ^1H NMR spectra of compounds, the signals of protons of the aromatic rings appeared in the region of 6.78 to 7.80 multiplet and 8.10 to 8.23 doublet for one proton in thiazole ring. The singlet due to the proton of C-H of methoxy group was observed at the *meta*-position and *para*-position 3.02 and 3.06 ppm, respectively and one singlet due to the proton of C-H of dimethyl amino group 2.97 ppm. In the ^{13}C NMR spectra of all compounds, carbons present in aromatic rings gave signals in the region of 112.1-149.6 ppm. The signal due to the CH_3-O carbon was observed at 56.0 ppm, whereas the signal due to the C-F carbon was observed at 163.10 ppm. The signals due to the CH_3-N and C-NO_2 carbons were observed at 40.8 ppm and 148.8 ppm, respectively. The signal due to the S-C-N carbon was observed at 152.6-153.1 ppm. The mass spectral data and elemental analysis were also complied with the structures of all the synthesized compounds.

Antimicrobial activity: The newly synthesized compounds **5a-j** were tested for *in vitro* antimicrobial activity. All synthesized compounds were evaluated against two Gram-positive bacteria, *Streptococcus aureus* and *Bacillus subtilis* and one Gram-negative bacteria, *Escherichia coli* with chloramphenicol serving as the reference compound and one fungi *Candida albicans* with ketoconazole serving as the reference compound. The activity was reported by measuring the diameter of inhibition zone (IZD) in mm and are depicted in Table-1. Almost all the tested compounds have shown moderate activity against all the tested bacteria and fungi. However, compounds **5b**, **5c** and **5g** showed good activity which is nearly equal to the inhibition activity of the standard bactericide and fungicide. The results indicated that the electron donating groups dimethyl amino group at position-4 (**5c**) and methoxy group in substi-

TABLE-1
PHYSICO-CHEMICAL, ANTIMICROBIAL AND CYTOTOXICITY DATA OF THE SYNTHESIZED IMIDAZO-THIAZOLES HYBRID DERIVATIVES (**5a-j**)

Comp. No.	R	Reaction time		Yield (%)		CTC_{50}[c] (µg/mL)	Diameter of zone of inhibition (mm) antimicrobial strains			
							Antibacterial			Antifungal
		MW[a] (min)	Conven[b] (h)	MW (min)	Conven (h)		Gram-positive		Gram-negative	*Candida albicans*
							S. aureus	*B. subtilis*	*E. coli*	
5a		11	11	85	52	110.70	5.1 (50)	6.2 (50)	8.2 (25)	11.7 (50)
5b		13	10	90	54	31.25	8.4 (25)	8.9 (12.5)	9.2 (12.5)	7.9 (12.5)
5c		14	13	88	51	100.21	9.8 (12.5)	8.1 (50)	9.2 (12.5)	9.2 (12.5)
5d		12	14	90	49	200.22	6.9 (50)	7.1 (50)	8.2 (25)	11.8 (25)

5e		12	14	87	55	91.25	11.9 (25)	11.2 (50)	8.2 (25)	11.8 (50)
5f		15	12	82	51	51.61	11.8 (50)	11.9 (25)	7.9 (12.5)	10.5 (50)
5g		14	16	89	60	44.41	10.1 (25)	9.2 (12.5)	8.2 (12.5)	8.2 (12.5)
5h		12	16	88	51	205.50	8.5 (50)	10.2 (100)	10.6 (50)	10.6 (100)
5i		15	14	82	49	91.61	9.8 (50)	9.6 (25)	8.2 (25)	9.5 (100)
5j		14	12	89	51	31.25	9.6 (50)	9.1 (100)	8.2 (25)	9.6 (50)

[a]MW: Microwave irradiation, [b]Conven: Conventional, [c]CTC_{50} : Cytotoxic Concentration (Which inhibited 50 % of total cells)

tuted phenyl group at position 3 & 4 (**5b** and **5g)** are due to the enhanced antibacterial and antifungal activities.

Anticancer activity: All synthesized compounds were screened for *in vitro* anticancer activity using DLA cell lines. The CTC_{50} value for all the synthesized compounds is also shown in Table-1. Compounds **5b**, **5f**, **5g** and **5j** showed the moderate anticancer activity at CTC_{50} values 31.25, 51.61, 44.41 and 31.25 µg/mL, respectively under *in vitro* anticancer screening using DLA cell lines in comparison to standard 5-fluorouracil showing CTC_{50} value at 31.25 µg/mL. Compounds **5f** and **5j** containing nitro group, which is electronegative in nature, make the compound potent. Similarly, the presence of methoxy group in compounds **5b** and **5g** also increases the cytotoxicity.

Antioxidant activity: All the newly synthesized imidazo-thiazole dihybrid derivatives were screened for free radical scavenging activity by DPPH method. Sample solutions were prepared to have concentrations of 10, 20, 30, 40 and 50 µg/mL. Ascorbic acid was taken as a standard antioxidant. Compounds **5a**, **5d**, **5e**, **6f**, **5i** and **5j** were found to be good free radical scavengers with good percentage inhibition in all the studied concentrations. The percentage inhibition for all the synthesized compounds is shown in Table-2. According to the results, the imidazo-thiazole hybrid derivatives containing substituted aldehyde having electronwithdrawing groups were found to have good antioxidant activities. Compounds **5a** (2,6-dichlorophenyl), **5d** (4-chlorophenyl), **5e** (4-fluorophenyl), **5f** (4-nitrophenyl), **5i** (3-chlorophenyl) and **5j** (3-nitrophenyl) have good potential as an antioxidant due to the presence of electronegative groups and thus enhanced the antioxidant activity.

Conclusion

A new series of hybrid analogous of imidazo-thiazole derivatives (**5a-j**) was synthesized and characterized with elemental analysis, ^{1}H NMR, ^{13}C NMR, FTIR, mass spectrophotometric techniques. The synthsized compounds were also

TABLE-2
DPPH FREE RADICAL SCAVENGING ACTIVITY OF THE SYNTHESIZED IMIDAZO-THIAZOLES HYBRID DERIVATIVES (**5a-j**)

Compounds	% Inhibition at µg/mL*					IC_{50} value (µmol/L)
	10	20	30	40	50	
5a	50.19 ± 1.05	54.71 ± 0.59	65.45 ± 0.12	73.49 ± 0.81	76.70 ± 0.71	25.18
5b	1.88 ± 0.19	7.67 ± 1.52	16.98 ± 0.51	20.37 ± 0.51	26.03 ± 0.54	220.12
5c	4.90 ± 0.61	8.30 ± 0.61	13.20 ± 0.51	20.75 ± 0.45	24.90 ± 0.61	219.88
5d	44.67 ± 0.17	50.00 ± 0.68	56.62 ± 0.71	59.83 ± 0.55	67.46 ± 0.64	44.22
5e	47.04 ± 0.57	53.58 ± 1.13	57.10 ± 0.57	63.39 ± 0.75	74.08 ± 0.57	35.61
5f	50.59 ± 1.17	53.61 ± 0.79	57.02 ± 0.62	60.94 ± 0.45	67.66 ± 0.75	28.09
5g	7.42 ± 0.75	15.66 ± 0.30	20.88 ± 0.62	30.82 ± 0.45	35.13 ± 0.75	174.22
5h	2.26 ± 0.37	8.55 ± 0.94	15.34 ± 0.78	20.00 ± 0.75	28.17 ± 0.94	213.82
5i	43.52 ± 0.57	54.00 ± 1.15	59.11 ± 0.57	66.03 ± 0.75	72.83 ± 1.13	44.47
5j	44.90 ± 0.37	54.08 ± 0.57	60.37 ± 0.37	65.15 ± 0.67	76.10 ± 0.62	39.46
Ascorbic acid	44.44 ± 1.23	55.12 ± 0.58	62.60 ± 0.63	66.81 ± 0.70	78.01 ± 0.53	87.67

*n = 3 (results are average of triplicate readings with standard deviation)

evaluated for their antimicrobial, anticancer and antioxidant activities. Compounds **5b**, **5c** and **5g** were found to be the best antibacterial action, whereas the cytotoxicity studies showed that compounds **5b**, **5f**, **5g** and **5j** have shown the best anticancer action against DLA cell lines. Compounds **5a**, **5d**, **5e**, **6f**, **5i** and **5j** were found to be the best free radical scavengers. Based on the results, the conclusion can be made that the compounds having substituted phenyl (dimethyl amino and methoxy) group possess for antibacterial and antifungal activities and (nitro and methoxy) group possess good anticancer activity, while (chloro, fluoro and nitro) group on the 2-(substituted phenyl)-4,5-diphenyl-1-(thiazol-2-yl)-1*H*-imidazoles moiety possess good antioxidant activity.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

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