

Research Article

Green Route for Efficient Synthesis of Novel Amino Acid Schiff Bases as Potent Antibacterial and Antifungal Agents and Evaluation of Cytotoxic Effects

Harshita Sachdeva,¹ Rekha Saroj,¹ Sarita Khaturia,¹
Diksha Dwivedi,¹ and Om Prakash Chauhan²

¹ Department of Chemistry, Faculty of Engineering and Technology, Mody Institute of Technology and Science, Lakshmanagarh, Rajasthan 332311, India

² Maharshi Dayanand Saraswati University, Ajmer, Rajasthan 305009, India

Correspondence should be addressed to Harshita Sachdeva; drhmsachdevaster@gmail.com

Received 7 May 2013; Revised 2 November 2013; Accepted 20 November 2013; Published 30 January 2014

Academic Editor: Marijan Kocevar

Copyright © 2014 Harshita Sachdeva et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Green chemical one-pot multicomponent condensation reaction of substituted 1*H*-indole-2,3-diones (**1**), various amino acids (**2**), and thiosemicarbazide (**3**) is found to be catalyzed by lemon juice as natural acid using water as a green solvent to give the corresponding Schiff bases (**4**) in good to excellent yields. This method is experimentally simple, clean, high yielding, and green, with reduced reaction times. The product is purified by simple filtration followed by washing with water and drying process. The synthesized compounds are characterized by FT-IR, ¹³CNMR, and ¹HNMR spectroscopy and are screened for their antifungal activity against *Aspergillus niger*, *Penicillium notatum*, *Fusarium oxysporum*, *Alternaria brassicicola*, *Chaetomium orium*, and *Lycopodium sp.* and antibacterial activity against Gram-positive bacteria, *Bacillus licheniformis*, *Staphylococcus aureus*, and *Micrococcus luteus*, and Gram-negative bacteria, *Pseudomonas aeruginosa* and *Escherichia coli*. Compounds have also been evaluated for cytotoxic effects against human colon cancer cell line Colo205.

1. Introduction

In recent years, environmentally benign synthetic methods have received considerable attention and some green protocols have been developed [1, 2]. There has been a large emphasis both in the chemical industry and in academic research on the development of environmentally benign solvents and reaction conditions. Organic synthetic procedures use organic solvents like benzene and chlorinated hydrocarbons, which have created havoc to the environment because of their toxic and volatile nature [3, 4].

To reduce such disasters there is a need to use a safer reaction medium like water. Water is nontoxic, safe, and cheap and never leads to menace to the environment [5–10]. The use of water as a solvent [11–13] is undoubtedly the best alternative as there are generally no harsh reaction conditions and no need of vigorous drying of the solvents.

Schiff bases have been playing vital roles in the preparation of a large number of industrial and biologically active compounds *via* closure, cycloaddition, and replacement reactions. They are also known for their diversified biological activities such as anti-inflammatory, allergic inhibitors, radical scavenging, analgesic and antioxidative action, and so forth [14–16]. Further, the incorporation of oxindole nuclei is an important synthetic strategy in drug discovery. Oxindoles have shown an extensive range of biological effects, including antibacterial, antifungal, anticonvulsant, antiviral, and antiproliferative activity [17]. On the other hand, heterocyclic compounds possessing 1,3,4-thiadiazole ring system show antifungal, bacteriostatic, anthelmintic [18] effect as well as depression effect on central nervous system [19]. Palaska et al. [20] reported 1,3,4-thiadiazole derivatives that exhibited analgesic and anti-inflammatory activities.

Considering the numerous applications of Schiff bases in various fields of chemistry, there has been tremendous interest in developing efficient methods for their preparation. Synthesis of Schiff base is often carried out with acid catalyst and generally by refluxing the mixture of aldehyde (or ketone) and amine in organic medium [21]. Classical organic synthesis of Schiff bases commonly meets the problem of removing solvents from the reaction mixture [22]. To overcome the difficulties in the removal of water, alternative method has been employed in which Lewis acid is used as catalyst which accelerates nucleophilic attack of amines on carbonyl carbon as well as serving as dehydrating agent for removal of the water. Several methods have been reported in the literature in which Lewis acids were used as catalysts such as $ZnCl_2$ [23], $TiCl_4$ [24], alumina [25], and P_2O_5 [26] and also by using materials like hydrotalcite [27]. Environmentally benign methods for the synthesis of Schiff bases have also been reported in the literature [28–32]. Although reported methodologies have their own advantages, they are overled by prolonged reaction time, the high reaction temperatures, an excess of costly dehydrating reagents/catalysts, moisture sensitive catalysts, special apparatus, and so forth.

It has been observed that the incorporation of more than one bioactive heterocyclic moiety into a single framework may result in the production of novel heterocycles with enhanced bioactivity. Keeping in view the diverse biological activities associated with oxindole and thiaziazole, we have synthesized Schiff bases incorporating these moieties and the synthesized compounds were evaluated for their antibacterial, antifungal, and cytotoxic effects. Moreover, Schiff bases have been synthesized in aqueous medium under the framework of green chemistry in moderate to good yields and short reaction time. The condensation reaction occurs very efficiently in the presence of water and the product can be isolated simply by filtration without the need of any further purification.

2. Results and Discussion

In continuation of our work to develop green methodologies for the synthesis of organic compounds [33–36], herein we wish to report a mild and highly efficient procedure for the synthesis of 3-[1-(5-amino-[1,3,4]-thiadiazol-2-yl)substitutedethylimino]-5-substituted-2,3-dihydro-indol-2-one derivatives (**4**) using water as a green solvent and citric acid as a green catalyst (Scheme 1). **4** was synthesized by the one-pot multicomponent condensation reaction of substituted 1*H*-indole-2,3-diones (**1**), various amino acids (**2**), and thiosemicarbazide (**3**) in the presence of lemon juice. The role of lemon juice in catalyzing the reaction was demonstrated by the lack of Schiff base formation when the reaction was carried out in the absence of catalyst. As lemon juice is acidic in nature ($pH \approx 2-3$) and percentage of citric acid (5–7%) is more than other acids, it works as acid catalyst for Schiff base formation [37]. Using this methodology these reactions were completed in shorter reaction times (2–3 hrs) with yields of the product ranging from 70 to 86%. For the present reaction, we have used extract of *Citrus limonum*

species of lemon as natural catalyst. The main ingredients of lemon juice are moisture (85%), carbohydrates (11.2%), citric acid (5–7%), protein (1%), vitamin C (0.5%), fat (0.9%), minerals (0.3%), fibers (1.6%), and some other organic acids.

This green route method requires simple work-up procedure, that is, simple filtration to isolate the products as they are insoluble in water and the desired products are obtained with satisfactory yields without any further purification. Considering the reaction time with water as solvent and yield of products this process was selected as green, environmental benign, clean, and safe to promote the synthesis of various Schiff's bases (Table 1). This acceleration has been attributed to many factors, including the hydrophobic effect [38, 39], enhanced hydrogen bonding in the transition state [40], and the high cohesive energy density of water [41, 42].

Initially, we attempted to synthesize Schiff bases incorporating oxindole and 1,3,4-thiadiazole moieties in water by the reaction of **1**, **2**, and **3** in the presence of $POCl_3$. Reaction gets completed in 5–6 hrs resulting in the formation of **4** with yields ranging from 60 to 70%. After the successful completion of reaction in water in the presence of $POCl_3$, we decided to synthesize Schiff bases by employing lemon juice (2 mL) as green catalyst for green approach (Scheme 1) resulting in quantitative yield (70–86%) of the corresponding Schiff bases within 2 to 3 hrs. However, in the absence of lemon juice, spiro(indole-triazoles) are formed [43]. We have also carried out this reaction in the presence of citric acid but the yield of the product (**4**) was found to be low (60–70%) as compared to that carried out using lemon juice as a green catalyst (70–86%). All the synthesized compounds were found to be chiral [44]. The occurrence of E/Z isomerism at the imine band of the molecules was suggested by Sari and Gürkan [45].

To establish the scope and limitations of lemon juice as a catalyst for Schiff base formation, structurally diverse 1*H*-indole-2,3-diones were treated with variously substituted amino acids under the catalytic influence of lemon juice and the results are summarized in Table 1. The purity of the compounds was checked by TLC using silica gel-G as adsorbent. The identity of the products obtained was confirmed by their IR, ^{13}C NMR, and 1H NMR spectral studies. The stretching bands derived from $-OH$ and $C=O$ of carboxylic acid were disappeared with the appearance of new two bands at 3259 and 3157 cm^{-1} due to asymmetric and symmetric stretching vibrations of NH_2 group. This evidence clearly confirmed the formation of compound **4**.

In summary, it can be stated that the present green synthetic protocol is highly efficient as it avoids the use of hazardous solvents at any stage of the reaction. As cited in Scheme 1, not only does the use of water as solvent allow for rapid reactions, but also the products are often insoluble in water, facilitating their ready isolation.

3. Experimental Section

3.1. General. Reagents and solvents were obtained from commercial sources and used without further purification. Melting points were determined on a Toshniwal apparatus.

TABLE 1: Results of synthesis of **4a-p** in the presence of lemon juice as natural acid.

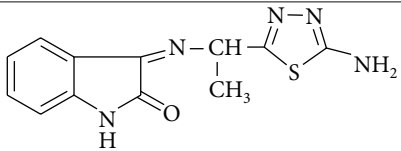
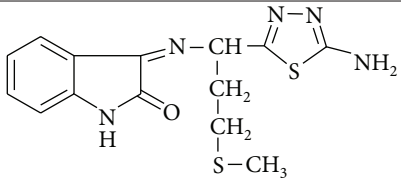
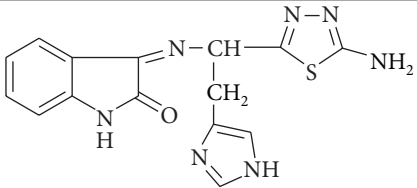
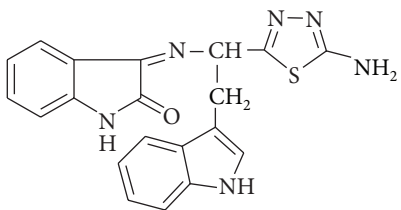
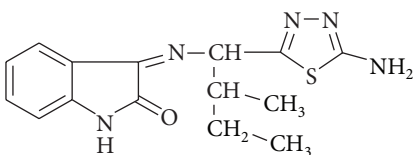
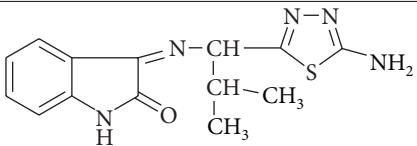
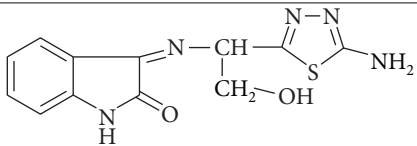
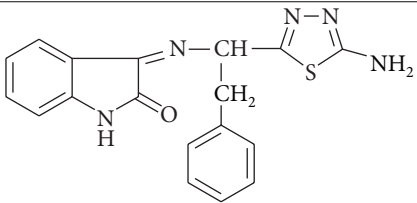
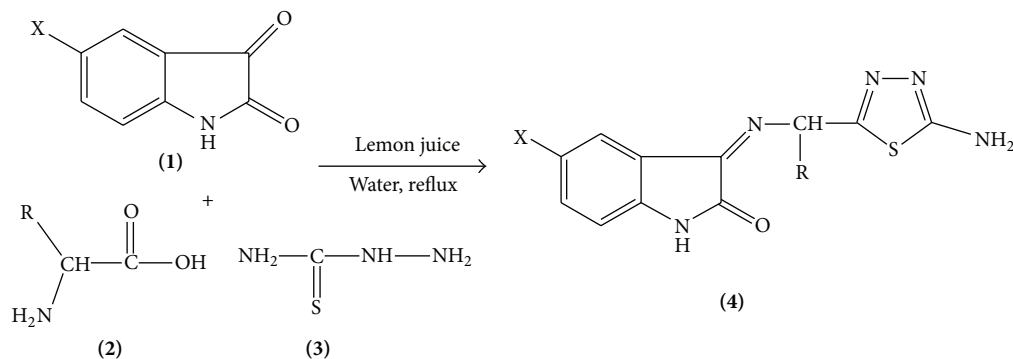
Entry	Product	Yield (%)	M.P. (°C)	Time (hrs)	Color
4a		86	195	2-3	Brown
4b		74	170	2-3	Brown
4c		78	103	2-3	Dark brown
4d		75	190	2-3	Dark brown
4e		74	170	2-3	Brown
4f		77	190	2-3	Brown
4g		70	98	2-3	Brown
4h		72	140	2-3	Red

TABLE I: Continued.

Entry	Product	Yield (%)	M.P. (°C)	Time (hrs)	Color
4i		70	140	2-3	Yellow
4j		72	202	2-3	Orange
4k		77	190	2-3	Red
4l		73	180	2-3	Red
4m		85	210	2-3	Yellow
4n		76	203	2-3	Yellow
4o		72	130	2-3	Pale yellow
4p		74	212	2-3	Pale yellow



SCHEME 1

The spectral analyses of synthesized compounds have been carried out at SAIF, Punjab University, Chandigarh. Purity of all compounds was checked by TLC using “G” coated glass plates and n-hexane: ethyl acetate (7:3) as eluent. IR spectra were recorded in KBr on a Perkin Elmer Infrared RXI FTIR spectrophotometer and ^1H NMR spectra were recorded on Bruker Avance II 400 NMR spectrometer using DMSO-d_6 and CDCl_3 as solvent and tetramethylsilane (TMS) as internal reference standard. The obtained products were identified from their spectral (^1H NMR, ^{13}C NMR, and IR) studies.

3.2. General Procedure for Extraction of Lemon Juice. Fresh lemon was cut by using knife and then pieces were pressed manually using domestic presser to extract juice. Then juice was then filtered through cotton/muslin cloth and then through filter paper to remove solid material and to get clear juice which was used as a catalyst.

3.3. Green Route for the Synthesis of 3-[1-(5-Amino-[1,3,4]-thiadiazol-2-yl)-substitutedethylimino]-5-substituted-2,3-dihydro-indol-2-ones (4a-p). An equimolar mixture of substituted 1H-indole-2,3-diones (0.012 mol), amino acids (0.012 mol), and thiosemicarbazide in water (15 mL containing 1 mL of ethanol) in the presence of lemon juice (2 mL) was mixed in round bottom flask and the mixture was refluxed for the time needed to complete the reaction (as monitored by TLC). The initial syrupy reaction mixture solidifies within 2-3 hrs. After completion of reaction, mixture was cooled to room temperature and the solid mass obtained was filtered, washed with water, and dried. The desired products were obtained with satisfactory yields without any further purification.

The spectroscopic characterization data of (4a-p) are given below.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-ethylimino]-2,3-dihydro-indol-2-one (4a). m.p. 195°C; IR (KBr): 3259, 3157, 2923, 1706, 1619, 1585, 1462, 673 cm^{-1} ; ^1H NMR (DMSO-d_6): 1.56 (d, 3H, CH_3), 4.59 (q, 1H, CH), 7.03–7.67 (m, 4H, aromatic), 7.22

(s, 2H, NH_2), 11.18 (s, 1H, N–H) ppm. ^{13}C -NMR (400 MHz, DMSO): 23.8, 51.4, 120, 122, 124, 128, 132, 146.4, 162, 176, 178. Anal.calcd for $\text{C}_{12}\text{H}_{11}\text{N}_5\text{OS}$: C, 52.73; H, 4.06; N, 25.62. Found: C, 52.91; H, 4.09; N, 25.63.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-3-methylsulfanyl-propylimino]-2,3-dihydro-indol-2-one (4b). m.p. 170°C; IR (KBr): 3263, 3177, 2980, 1706, 1620, 1580, 1462, 670 cm^{-1} ; ^1H NMR (DMSO-d_6): 2.09 (s, 3H, CH_3), 2.31 (q, 2H, CH_2), 2.44 (t, 2H, CH_2), 4.41 (t, 1H, CH), 7.03–7.67 (m, 4H, aromatic), 7.22 (s, 2H, NH_2), 11.18 (s, 1H, N–H) ppm. ^{13}C -NMR (400 MHz, DMSO): 17, 30.4, 38.2, 52.4, 122, 124, 126, 128, 132, 146.2, 160, 172, 174. Anal.calcd for $\text{C}_{14}\text{H}_{15}\text{N}_5\text{OS}_2$: C, 50.43; H, 4.53; N, 21.00. Found: C, 50.24; H, 4.56; N, 21.02.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-(1H-imidazol-4-yl)-ethylimino]-2,3-dihydro-indol-2-one (4c). m.p. 103°C; IR (KBr): 3260, 3175, 2986, 1705, 1619, 1585, 1462, 673 cm^{-1} ; ^1H NMR (DMSO-d_6): 3.33 (d, 2H, CH_2), 4.80 (t, 1H, CH), 6.80 (d, 1H, CH), 8.73 (d, 1H, CH), 7.03–7.67 (m, 4H, aromatic), 7.22 (s, 2H, NH_2), 11.18 (s, 1H, N–H), 13.4 (dd, 1H, NH) ppm. ^{13}C -NMR (400 MHz, DMSO): 36, 56, 118, 120, 122.6, 124.8, 128.2, 132.2, 134.2, 136.8, 146.8, 162.8, 174.2, 176. Anal.calcd for $\text{C}_{15}\text{H}_{13}\text{N}_7\text{OS}$: C, 53.09; H, 3.86; N, 28.89. Found: C, 53.27; H, 3.89; N, 28.91.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-(1H-indol-3-yl)-ethylimino]-2,3-dihydro-indol-2-one (4d). m.p. 190°C; IR (KBr): 3263, 3177, 2980, 1706, 1619, 1580, 1462, 670 cm^{-1} ; ^1H NMR (DMSO-d_6): 3.20 (d, 2H, CH_2), 4.80 (t, 1H, CH), 7.18 (s 1H, CH), 10.85 (s, 1H, NH), 7.03–7.67 (m, 4H, aromatic), 6.97–7.58 (m, 4H, aromatic), 7.22 (s, 2H, NH_2), 11.18 (s, 1H, N–H) ppm. ^{13}C -NMR (400 MHz, DMSO): 36.8, 58, 110, 112, 118, 120.2, 122.8, 124, 128, 130.2, 132.6, 136, 146, 164.4, 176, 178.2. Anal.calcd for $\text{C}_{20}\text{H}_{16}\text{N}_6\text{OS}$: C, 61.84; H, 4.15; N, 21.63. Found: C, 61.66; H, 4.18; N, 21.65.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-methylbutylimino]-2,3-dihydro-indol-2-one (4e). m.p. 170°C; IR (KBr): 3263, 3086, 2986, 2876, 1733, 1621, 1594, 1462, 673 cm^{-1} ; ^1H NMR

(DMSO- d_6): 0.96 (t, 3H, CH₃), 1.29 (q, 2H, CH₂), 1.06 (d, 3H, CH₃), 2.00 (m, 1H, CH), 4.40 (d, 1H, CH), 7.03–7.67 (m, 4H, aromatic), 7.22 (s, 2H, NH₂), 11.18 (s, 1H, N–H) ppm. ¹³C-NMR (400 MHz, DMSO): 11.6, 15.8, 24.8, 38, 58.2, 120.2, 122.2, 124.8, 128, 132, 142.2, 162.2, 176.2, 178. Anal.calcd for C₁₅H₁₇N₅OS: C, 57.12; H, 5.43; N, 22.21. Found: C, 57.32; H, 5.45; N, 22.23.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-methylpropylimino]-2,3-dihydro-indol-2-one (**4f**). m.p. 190°C; IR (KBr): 3260, 3150, 2877, 2803, 1695, 1620, 1580, 1465, 670 cm⁻¹; ¹H NMR (DMSO- d_6): 1.01 (d, 3H, CH₃), 1.01 (d, 3H, CH₃), 2.2 (m, 1H, CH), 4.40 (d, 1H, CH), 7.03–7.67 (m, 4H, aromatic), 7.22 (s, 2H, NH₂), 11.18 (s, 1H, N–H) ppm. ¹³C-NMR (400 MHz, DMSO): 18.2, 36.2, 78.6, 110, 126, 128, 130, 142, 143, 162.8, 176, 178.2. Anal.calcd for C₁₄H₁₅N₅OS: C, 55.80; H, 5.02; N, 23.24. Found: C, 55.62; H, 5.04; N, 23.25.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-hydroxyethylimino]-2,3-dihydro-indol-2-one (**4g**). m.p. 98°C; IR (KBr): 3465, 3263, 3177, 2980, 1706, 1619, 1580, 1462, 673 cm⁻¹; ¹H NMR (DMSO- d_6): 4.30 (d, 2H, CH₂), 4.50 (t, 1H, CH), 4.78 (s, 1H, OH), 7.03–7.67 (m, 4H, aromatic), 7.22 (s, 2H, NH₂), 11.18 (s, 1H, N–H) ppm. ¹³C-NMR (400 MHz, DMSO): 51.4, 68, 120, 122, 124, 128, 132, 146.2, 162.8, 176.2, 178.8. Anal.calcd for C₁₂H₁₁N₅O₂S: C, 49.82; H, 3.83; N, 24.21. Found: C, 49.63; H, 3.85; N, 24.22.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-phenylethylimino]-2,3-dihydro-indol-2-one (**4h**). m.p. 140°C; IR (KBr): 3265, 3175, 2986, 1705, 1620, 1580, 1465, 670 cm⁻¹; ¹H NMR (DMSO- d_6): 3.33 (d, 2H, CH₂), 4.80 (t, 1H, CH), 7.12–7.26 (m, 5H, aromatic), 7.03–7.67 (m, 4H, aromatic), 7.22 (s, 2H, NH₂), 11.18 (s, 1H, N–H) ppm. ¹³C-NMR (400 MHz, DMSO): 42.8, 56.4, 120, 122, 124.2, 128.8, 132, 138, 146.2, 162.8, 176.2, 178. Anal.calcd for C₁₈H₁₅N₅OS: C, 61.87; H, 4.33; N, 20.04. Found: C, 61.68; H, 4.35; N, 20.06.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-phenylethylimino]-5-chloro-2,3-dihydro-indol-2-one (**4i**). m.p. 140°C; IR (KBr): 3263, 3177, 2980, 1706, 1619, 1580, 1462, 695, 670 cm⁻¹; ¹H NMR (DMSO- d_6): 3.33 (d, 2H, CH₂), 4.80 (t, 1H, CH), 7.12–7.26 (m, 5H, aromatic), 7.28–7.61 (m, 3H, aromatic), 7.22 (s, 2H, NH₂), 11.18 (s, 1H, N–H) ppm. ¹³C-NMR (400 MHz, DMSO): 42, 56, 120.2, 122.4, 124, 128, 132, 144, 162, 176, 178.8. Anal.calcd for C₁₈H₁₄ClN₅OS: C, 56.32; H, 3.68; N, 18.24. Found: C, 56.51; H, 3.66; N, 18.25.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-(1H-imidazol-4-yl)ethylimino]-5-chloro-2,3-dihydro-indol-2-one (**4j**). m.p. 202°C; IR (KBr): 3265, 3170, 2985, 1705, 1619, 1580, 1462, 698, 670 cm⁻¹; ¹H NMR (DMSO- d_6): 3.33 (d, 2H, CH₂), 4.80 (t, 1H, CH), 6.80 (d, 1H, CH), 8.73 (d, 1H, CH), 7.28–7.61 (m, 3H, aromatic), 7.22 (s, 2H, NH₂), 11.18 (s, 1H, N–H), 13.4 (dd, 1H, NH) ppm. ¹³C-NMR (400 MHz, DMSO): 38, 56.8, 120, 122.2, 124, 128.6, 132, 136, 162.2, 176.4, 178. Anal.calcd for C₁₅H₁₂ClN₇OS: C, 48.19; H, 3.24; N, 26.23. Found: C, 48.38; H, 3.26; N, 26.24.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-3-methylsulfanyl-propylimino]-5-chloro-2,3-dihydro-indol-2-one (**4k**). m.p. 190°C; IR (KBr): 3263, 3177, 2980, 1706, 1620, 1580, 1465, 690, 673 cm⁻¹; ¹H NMR (DMSO- d_6): 2.09 (s, 3H, CH₃), 2.31 (q, 2H, CH₂), 2.44 (t, 2H, CH₂), 4.41 (t, 1H, CH), 7.28–7.61 (m, 3H, aromatic), 7.22 (s, 2H, NH₂), 11.18 (s, 1H, N–H) ppm. ¹³C-NMR (400 MHz, DMSO): 18, 30.4, 38, 54.4, 120.2, 122.8, 124.4, 128, 132, 144, 162, 176 and 178. Anal.calcd for C₁₄H₁₄ClN₅OS₂: C, 45.71; H, 3.84; N, 19.04. Found: C, 45.52; H, 3.86; N, 19.05.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-(1H-indol-3-yl)ethylimino]-5-chloro-2,3-dihydro-indol-2-one (**4l**). m.p. 180°C; IR (KBr): 3265, 3175, 2986, 1705, 1619, 1580, 1462, 695, 670 cm⁻¹; ¹H NMR (DMSO- d_6): 3.20 (d, 2H, CH₂), 4.80 (t, 1H, CH), 7.18 (s, 1H, CH), 10.85 (s, 1H, NH), 7.28–7.61 (m, 3H, aromatic), 6.97–7.58 (m, 4H, aromatic), 7.22 (s, 2H, NH₂), 11.18 (s, 1H, N–H) ppm. ¹³C-NMR (400 MHz, DMSO): 36, 58.4, 110, 112, 120, 122, 124, 128.2, 132, 136, 144.4, 162.8, 176.2, 178.2. Anal.calcd for C₂₀H₁₅ClN₆OS: C, 56.80; H, 3.58; N, 19.87. Found: C, 56.61; H, 3.55; N, 19.89.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-phenylethylimino]-5-nitro-2,3-dihydro-indol-2-one (**4m**). m.p. 210°C; IR (KBr): 3263, 3177, 2980, 1706, 1615, 1580, 1462, 1362, 670 cm⁻¹; ¹H NMR (DMSO- d_6): 3.33 (d, 2H, CH₂), 4.80 (t, 1H, CH), 7.12–7.26 (m, 5H, aromatic), 7.93–8.53 (m, 3H, aromatic), 7.22 (s, 2H, NH₂), 11.18 (s, 1H, N–H) ppm. ¹³C-NMR (400 MHz, DMSO): 42, 56.4, 120, 122, 124, 128, 132, 144, 152, 162, 176, 178. Anal.calcd for C₁₈H₁₄N₆O₃S: C, 54.81; H, 3.58; N, 21.31. Found: C, 54.62; H, 3.56; N, 21.34.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-(1H-imidazol-4-yl)ethylimino]-5-nitro-2,3-dihydro-indol-2-one (**4n**). m.p. 203°C; IR (KBr): 3265, 3175, 2986, 1705, 1619, 1585, 1460, 1365, 672 cm⁻¹; ¹H NMR (DMSO- d_6): 3.33 (d, 2H, CH₂), 4.80 (t, 1H, CH), 6.80 (d, 1H, CH), 8.73 (d, 1H, CH), 7.93–8.53 (m, 3H, aromatic), 7.22 (s, 2H, NH₂), 11.18 (s, 1H, N–H), 13.4 (dd, 1H, NH) ppm. ¹³C-NMR (400 MHz, DMSO): 37, 56, 118, 120.2, 122.8, 124.6, 128.2, 132.2, 144.4, 152.2, 162.8, 176.2, 178.4. Anal.calcd for C₁₅H₁₂N₈O₃S: C, 46.87; H, 3.15; N, 29.15. Found: C, 46.68; H, 3.17; N, 29.17.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-3-methylsulfanyl-propylimino]-5-nitro-2,3-dihydro-indol-2-one (**4o**). m.p. 130°C; IR (KBr): 3260, 3170, 2985, 1702, 1615, 1580, 1462, 1362, 670 cm⁻¹; ¹H NMR (DMSO- d_6): 2.09 (s, 3H, CH₃), 2.31 (q, 2H, CH₂), 2.44 (t, 2H, CH₂), 4.41 (t, 1H, CH), 7.93–8.53 (m, 3H, aromatic), 7.22 (s, 2H, NH₂), 11.18 (s, 1H, N–H) ppm. ¹³C-NMR (400 MHz, DMSO): 18, 30, 38, 54, 122.2, 124.8, 128.4, 134.2, 144.8, 152.6, 162.2, 176.4, 178.2. Anal.calcd for C₁₄H₁₄N₆O₃S₂: C, 44.43; H, 3.73; N, 22.21. Found: C, 44.62; H, 3.71; N, 22.23.

3-[1-(5-Amino-[1,3,4]thiadiazol-2-yl)-2-(1H-indol-3-yl)ethylimino]-5-nitro-2,3-dihydro-indol-2-one (**4p**). m.p. 212°C; IR (KBr): 3263, 3177, 2980, 1706, 1620, 1585, 1460, 1362, 675 cm⁻¹; ¹H NMR (DMSO- d_6): 3.20 (d, 2H, CH₂), 4.80 (t, 1H, CH), 7.18 (s, 1H, CH), 10.85 (s, 1H, NH), 7.93–8.53 (m, 3H, aromatic), 6.97–7.58 (m, 4H, aromatic), 7.22 (s, 2H, NH₂),

11.18 (s, 1H, N-H) ppm. ^{13}C -NMR (400 MHz, DMSO): 36, 58.4, 110, 118, 120.2, 122, 124, 128, 132, 144, 152, 162, 176.8, 178. Anal. calcd for $\text{C}_{20}\text{H}_{15}\text{N}_7\text{O}_3\text{S}$: C, 55.42; H, 3.49; N, 22.62. Found: C, 55.23; H, 3.47; N, 22.63.

4. Antimicrobial Activity

4.1. Antibacterial Activity. All the synthesized compounds were tested for in vitro antibacterial activity by twofold serial dilution assay [46] against the reference compound streptomycin. The minimal inhibitory concentration (MIC) is shown in Tables 2 and 4. During MIC determination by twofold serial dilution method, compounds **4g**, **4j**, and **4m** exhibit good activities against all bacterial strains at a MIC value of 1000 $\mu\text{g}/\text{mL}$. Admirable activities against *M. luteus* and *E. coli* are displayed by compounds **4d**, **4e**, **4g**, **4h**, **4i**, **4j**, **4l**, and **4o** at a MIC value of 500 $\mu\text{g}/\text{mL}$. Compounds, **4a**, **4e**, **4o**, and **4p** display higher activities against *S. aureus*, at a MIC value of 1000 $\mu\text{g}/\text{mL}$.

4.2. Antifungal Activity. The antifungal activity of the compounds was also studied with *Aspergillus sp.*, *Penicillium sp.*, *Alternaria brassicicola*, *Chaetomium orium*, and *Lycopodium sp. fungi*. The results are summarized in Tables 2 and 3. It has been observed that all synthesized chemical compounds showed lesser antifungal activity as compared to antibacterial activity. On MIC determination, compounds **4h**, **4i**, **4k**, and **4m** showed excellent antimicrobial activities at a MIC value of 1000 $\mu\text{g}/\text{mL}$. Modest activities are displayed by compound **4n** against *Aspergillus sp.* and *Chaetomium orium* at a MIC value of >1000 $\mu\text{g}/\text{mL}$, whereas compound **4a**, **4c**, and **4g** exhibit mild activities against *Aspergillus sp.* and *Lycopodium sp.* at a MIC value of 1000 $\mu\text{g}/\text{mL}$.

Determination of Minimal Inhibitory Concentrations (MIC). Minimum inhibitory concentration was determined by twofold serial dilution method [46]. A series of test tubes were prepared containing the same volume of media inoculated with the test organism. Drug was added to the tubes in a stepwise dilution by a factor of 2 (twofold serial dilution); that is, the concentration of drug in the first tube is 1000 $\mu\text{g}/\text{mL}$, in the second tube it will be 500 $\mu\text{g}/\text{mL}$, and in the third it will be 250 $\mu\text{g}/\text{mL}$. Cultures were incubated at 24 hrs for bacteria at 37°C and 48 hrs for fungi at 30°C. One tube was left without drug only solvent to serve as a positive control for the growth of the organism. After incubation, tubes were examined for microbial growth and the minimum inhibitory concentrations (the minimum concentration of each treatment that inhibits microbial growth after the incubation period) were measured. Tubes are inspected visually to determine the growth of the organism indicated by turbidity and simultaneously streak on Petri dish containing nutrient agar medium; after incubation (24 hours for bacteria and 48 hrs for fungal strains) microbial growth were measured.

From the screening results, it is observed that the presence of electron withdrawing groups like Cl and NO_2 group at 5th position of indole ring exhibits significant activity in comparison to unsubstituted compounds. However other

synthesized compounds of the series also exhibit moderate to significant activity against the microorganisms as mentioned above.

4.3. Evaluation of Cytotoxic Effects. The uncontrolled growth of cells in the body, started due to certain stimuli, lays the foundation of cancer. Anticancer drugs either kill cancer cells or modify their growth. Among the heterocyclic compounds, five membered heterocyclic moieties fused with aromatic ring system with various heteroatoms like N, S, and O are potential anticancer agents [47]. Synthesis of nitrogen containing heterocyclic compounds has been a subject of great interest due to the wide application in agrochemical and pharmaceutical fields. Heterocycles like indole, pyrimidine, pyridine, quinoline, and so forth are an integral part of a huge number of natural and synthetic compounds and play important roles in the biological systems. For developing the suitable leads for anticancer drugs, introduction of appropriate substituents at C-3 of indole is required [48]. It has also been observed that Schiff bases also play a major role in developing cytotoxic drugs [49, 50]. Hence, an attempt has been made to synthesize Schiff bases incorporating indole moiety and the synthesized compounds **4a**, **4e**, **4f**, and **4h** were evaluated [51] for their cytotoxic effects against Human colon cancer cell line Colo205. Percent growth was calculated on a plate-by-plate basis for test wells relative to control wells.

Percent growth was expressed as the ratio of average absorbance of the test well to the average absorbance of the control wells * 100.

Using the six-absorbance measurements [time zero (T_z), control growth (C), and test growth in the presence of drug at the four concentration levels (T_i)], the percentage growth was calculated at each of the drug concentration levels.

Percentage growth inhibition was calculated as

$$\left[\frac{(T_i - T_z)}{(C - T_z)} \right] \times 100 \text{ for concentrations for which } T_i \geq T_z \text{ (} T_i - T_z \text{) positive or zero,}$$

$$\left[\frac{(T_i - T_z)}{T_z} \right] \times 100 \text{ for concentrations for which } T_i < T_z. \text{ (} T_i - T_z \text{) negative.}$$

The dose response parameters were calculated for each test article. Growth inhibition of 50% (GI50) was calculated from $\left[\frac{(T_i - T_z)}{(C - T_z)} \right] \times 100 = 50$, which is the drug concentration resulting in a 50% reduction in the net protein increase (as measured by SRB staining) in control cells during the drug incubation. The drug concentration resulting in total growth inhibition (TGI) was calculated from $T_i = T_z$. The LC50 (concentration of drug resulting in a 50% reduction in the measured protein at the end of the drug treatment as compared to that at the beginning) indicating a net loss of cells following treatment is calculated from

$$\left[\frac{(T_i - T_z)}{T_z} \right] \times 100 = -50. \quad (1)$$

Values were calculated for each of these three parameters if the level of activity was reached; however, if the effect was not reached or was exceeded, the values for that parameter were expressed as greater or less than the maximum or minimum concentration tested. The data of anticancer

TABLE 3: Minimum inhibitory concentration for fungi.

Compound	MIC (ug/mL)														
	<i>Aspergillus</i> sp.			<i>Penicillium</i> sp.			<i>Alternaria brassicicola</i>			<i>Chaetomium orium</i>			<i>Lycopodium</i> sp.		
	250	500	1000	250	500	1000	250	500	1000	250	500	1000	250	500	1000
4a	+	+	+	+	+	+	+	+	+	+	+	+	+	-	--*
4b	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
4c	+	+	--*	+	+	+	+	+	+	+	+	+	+	+	+
4d	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
4e	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
4f	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
4g	+	+	-	+	+	+	+	+	+	+	+	+	-	+	+
4h	+	+	--*	+	+	+	+	+	--*	+	+	--*	+	+	+
4i	+	+	+	+	+	--*	+	+	--*	+	+	--*	+	+	+
4j	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
4k	+	+	+	+	+	--*	+	+	--*	+	+	--*	+	+	--*
4l	+	+	--*	+	+	+	+	+	+	+	+	-	+	+	+
4m	+	+	--*	+	+	--*	+	+	-	+	+	--*	+	+	-
4n	+	+	--*	+	+	+	+	+	-	+	+	--*	+	+	-
4o	+	+	+	+	+	+	+	+	-	+	+	+	+	+	-
4p	+	+	+	+	+	+	+	+	-	+	+	--*	+	+	--*
Solvent DMSO	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Itraconazole	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

*Moderately active (zone of inhibition 8–10 mm).

TABLE 4: Minimum inhibitory concentration for bacteria.

Compound	MIC (ug/mL)														
	<i>Pseudomonas aeruginosa</i>			<i>Bacillus licheniformis</i>			<i>Staphylococcus aureus</i>			<i>Micrococcus luteus</i>			<i>Escherichia coli</i>		
	250	500	1000	250	500	1000	250	500	1000	250	500	1000	250	500	1000
4a	+	+	+	+	+	+	+	-	--*	+	-	--*	+	-	--*
4b	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
4c	+	-	--*	+	-	--*	+	+	+	+	+	+	+	-	--*
4d	+	+	+	+	+	+	+	+	+	+	--*	--*	+	--*	--*
4e	+	+	+	+	+	+	+	-	--*	+	--*	--*	+	-	--*
4f	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
4g	+	-	--*	+	-	--*	+	-	--*	+	--*	--*	-	--*	--*
4h	+	+	+	+	-	--*	+	+	+	-	--*	--*	+	--*	--*
4i	+	+	+	+	-	--*	+	+	+	-	--*	--*	-	--*	--*
4j	+	-	--*	+	+	--*	+	+	--*	-	--*	--*	+	--*	--*
4k	+	+	+	+	+	+	+	+	+	+	+	+	+	-	--*
4l	+	+	--*	+	+	+	+	+	+	+	--*	--*	+	+	+
4m	+	-	--*	+	+	--*	+	-	--	+	+	+	+	-	--*
4n	+	+	-	+	+	--*	+	+	+	+	--*	--*	+	+	+
4o	+	+	-	+	+	+	+	-	--*	+	--*	--*	+	+	--*
4p	+	-	--*	+	+	--*	+	-	--*	+	+	+	+	+	+
Solvent DMSO	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Streptomycin	-	-	-	-	+	-	-	+	-	-	+	-	-	-	-
Gentamycin	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

+: inactive (zone of inhibition < 6 mm); -: slightly active (zone of inhibition 6–8 mm); --: moderately active (zone of inhibition 8–10 mm); - - - -: highly active (zone of inhibition > 10 mm); filled part showing MIC for particular organism.

TABLE 5: In vitro cytotoxic effects of amino acid Schiff bases against human colon cancer cell line Colo205.

(a)

Entry	Human colon cancer cell line Colo205															
	% control growth															
	Molar drug concentration															
	Experiment 1				Experiment 2				Experiment 3				Average values			
	10 ⁻⁶ M	10 ⁻⁵ M	10 ⁻⁴ M	10 ⁻³ M	10 ⁻⁷ M	10 ⁻⁶ M	10 ⁻⁵ M	10 ⁻⁴ M	10 ⁻⁷ M	10 ⁻⁶ M	10 ⁻⁵ M	10 ⁻⁴ M	10 ⁻⁷ M	10 ⁻⁶ M	10 ⁻⁵ M	10 ⁻⁴ M
4a	98.3	91.4	91.2	29.4	96.3	93.2	91.8	28.9	90.4	94.9	83.6	22.9	95.0	93.2	88.9	27.0
4e	100.0	100.0	100.0	23.0	100.0	100.0	100.0	23.7	100.0	100.0	100.0	23.8	100.0	100.0	100.0	23.5
4f	100.0	98.0	96.3	39.7	100.0	96.3	95.1	38.2	100.0	100.0	100.0	43.4	100.0	98.1	97.1	40.4
4h	100.0	100.0	93.1	-25.0	100.0	99.4	91.5	-20.5	100.0	97.9	88.1	-28.3	100.0	99.1	90.9	-24.6
ADR	73.4	21.7	-64.1	-74.7	71.1	23.3	-70.2	-73.5	67.5	14.1	-66.1	-76.3	70.7	19.7	-66.8	-74.8

(b)

Human colon cancer cell line Colo205				
Molar drug concentration calculated from graph				
Colo205	LC50	TGI	GI50	
4a	>100	>100	67.2	
4e	>100	>100	68.0	
4f	>100	>100	85.4	
4h	>100	81.6	41.8	
ADR	67.7	15.3	<10	

activity in terms of % control growth at different molar drug concentrations are shown in Table 4. Unfortunately, the evaluated compounds were not found to be active against human colon cancer cell line. Growth curve is presented in Figure 1.

4.3.1. Definitions and Notes

GI50: growth inhibition of 50% (GI50) is calculated from $[(Ti - Tz)/(C - Tz)] \times 100 = 50$, drug concentration resulting in a 50% reduction in the net protein increase;

TGI: drug concentration resulting in total growth inhibition (TGI) will be calculated from $Ti = Tz$;

LC50: concentration of drug resulting in a 50% reduction in the measured protein at the end of the drug treatment (as compared to that at the beginning) indicating a net loss of 50% cells following treatment is calculated from $[(Ti - Tz)/Tz] \times 100 = -50$;

ADR: Adriamycin (doxorubicin) is positive control compound;

GI50 value of $\leq 10^{-6}$ (i.e., 1 μ mole) or $\leq 10 \mu$ g/mL is considered to demonstrate activity in case of pure compounds.

For extracts, GI50 value $\leq 20 \mu$ g/mL is considered to demonstrate activity.

The bold highlighted test values under GI50 column indicate activity (see Table 5).

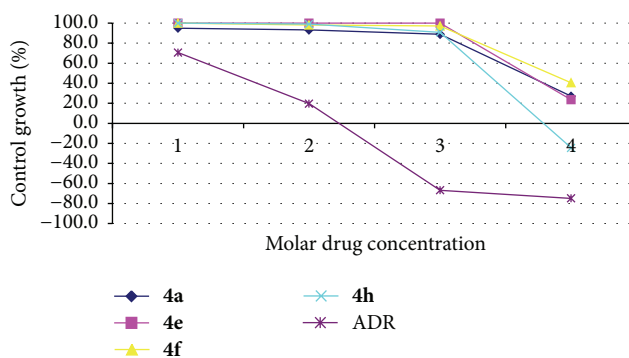


FIGURE 1: In vitro cytotoxic effects of amino acid Schiff bases against human colon cancer cell line Colo205.

5. Conclusions

We have developed a convenient, simple, efficient, and eco-friendly green procedure for the synthesis of Schiff bases from 1*H*-indole-2,3-diones, various amino acids, and thiosemicarbazide under mild reaction conditions in an aqueous medium. Some of the major advantages of this protocol are the ambient conditions, good to excellent yields, simple work-up procedure, and use of water as a desirable solvent for chemical reaction for reasons cost, safety, and environmental concerns. Further, use of lemon juice as green catalyst makes this methodology an alternative platform to organic solvent conventional synthesis under the umbrella of environmentally greener and safer processes. The synthesized compounds are found to be active against tested pathogens. The above

results of antifungal and antibacterial screening establish the fact that thiadiazole substituted indole derivatives can be studied further to explore out newer antimicrobial compounds.

Conflict of Interests

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

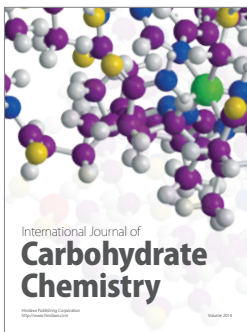
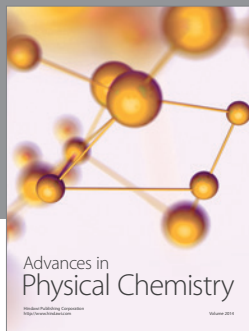
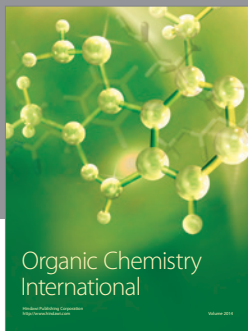
Acknowledgments

The authors are thankful to the Dean, FET, MITS, and HOD, Science and Humanities, for providing necessary research facilities in the department. Financial assistance from FET, MITS, is gratefully acknowledged. They are also thankful to the director, The Advanced Centre for Treatment, Research and Education in Cancer (ACTREC), TATA Memorial Centre, Mumbai, India, for anticancer screening, and SAIF, Punjab University, Chandigarh, India, for the spectral analyses of synthesized compounds.

References

- [1] V. K. Redasani, V. S. Kumawat, R. P. Kabra, P. Kansagara, and S. J. Surana, "Applications of green chemistry in organic synthesis," *International Journal of ChemTech Research*, vol. 2, no. 3, pp. 1856–1859, 2010.
- [2] S. Nishimura, A. Takagaki, and K. Ebitani, "Characterization, synthesis and catalysis of hydrotalcite-related materials for highly efficient materials transformations," *Green Chemistry*, vol. 15, no. 8, pp. 2026–2042, 2013.
- [3] M. Poliakoff, J. M. Fitzpatrick, T. R. Farren, and P. T. Anastas, "Green chemistry: science and politics of change," *Science*, vol. 297, no. 5582, pp. 807–810, 2002.
- [4] J. M. DeSimone, "Practical approaches to green solvents," *Science*, vol. 297, no. 5582, pp. 799–803, 2002.
- [5] C. J. Li and T. H. Chan, *Organic Reactions in Aqueous Media*, Wiley, New York, NY, USA, 1997.
- [6] G.-J. Ten Brink, I. W. C. E. Arends, and R. A. Sheldon, "Green, catalytic oxidation of alcohols in water," *Science*, vol. 287, no. 5458, pp. 1636–1639, 2000.
- [7] U. M. Lindström, "Stereoselective organic reactions in water," *Chemical Reviews*, vol. 102, no. 8, pp. 2751–2772, 2002.
- [8] A. M. Zonouz, I. Eskandari, and H. R. Khavasi, "A green and convenient approach for the synthesis of methyl-6-amino-5-cyano-4-aryl-2,4-dihydropyrano[2,3-c]pyrazole-3-carboxylates via a one-pot, multi-component reaction in water," *Tetrahedron Letters*, vol. 53, no. 41, pp. 5519–5522, 2012.
- [9] F. Aryanasab, A. Z. Halimehjani, and M. R. Saidi, "Dithiocarbamate as an efficient intermediate for the synthesis of 2-amino-1,3,4-thiadiazoles in water," *Tetrahedron Letters*, vol. 51, no. 5, pp. 790–792, 2010.
- [10] F. M. Moghaddam, M. Kiamehr, M. R. Khodabakhshi, Z. Mirjafary, S. Fathi, and H. Saeidian, "A new domino Knoevenagel-hetero-Diels-Alder reaction: an efficient catalyst-free synthesis of novel thiochromone-annulated thiopyranocoumarin derivatives in aqueous medium," *Tetrahedron*, vol. 66, no. 45, pp. 8615–8622, 2010.
- [11] S. S. Pawar, M. S. Shingare, and S. N. Thore, "A novel approach for ligand promoted palladium (II)-catalyzed Suzuki coupling of aryl iodides and bromides with arylboronic acid in aqueous media," *Letters in Organic Chemistry*, vol. 4, no. 7, pp. 486–490, 2007.
- [12] Y. Ren and C. Cai, "Iodine catalysis in aqueous medium: an improved reaction system for Knoevenagel and nitroaldol condensation," *Catalysis Letters*, vol. 118, no. 1-2, pp. 134–138, 2007.
- [13] V. K. Rao, S. S. Reddy, B. S. Krishna, K. R. M. Naidu, C. N. Raju, and S. K. Ghosh, "Synthesis of Schiff bases in aqueous medium: a green alternative approach with effective mass yield and high reaction rates," *Green Chemistry Letters and Reviews*, vol. 3, no. 3, pp. 217–223, 2010.
- [14] L. Hadjipavlu, J. Dimitra, Geronikaki, and A. Athina, "Thiazolyl and benzo-thiazolyl Schiff base as novel possible lipoxygenase inhibitors and anti-inflammatory agents," *Drug Design and Discovery*, vol. 15, pp. 199–206, 1998, Chemical Abstracts, vol.129, Article ID 148934, 1998.
- [15] B. De and G. V. S. Ramasarma, "Determination of PKa and correlation with the analgesic activity of 5-Oxoimidazolylaminopyrazole-4-carboxaldehydes and their Schiff's bases," *Indian Drugs*, vol. 36, no. 9, pp. 583–587, 1999.
- [16] X.-Y. Luo, J.-Z. Zhao, Y.-J. Lin, and Z.-Q. Liu, "Antioxidative effect of schiff bases with o-hydroxybenzylidene group on free radical induced hemolysis of human red blood cell," *Chemical Research in Chinese Universities*, vol. 18, no. 3, pp. 287–289, 2002, Chemical Abstracts, vol. 138, Article ID 247927, 2003.
- [17] J. F. M. da Silva, S. J. Garden, and A. C. Pinto, "The chemistry of isatins: a review from 1975 to 1999," *Journal of the Brazilian Chemical Society*, vol. 12, no. 3, pp. 273–324, 2001.
- [18] A. Marin, N. Valls, F. J. Berenguer et al., "Synthesis and antelmintic activity of carbamates derived from imidazo [2, 1-b][1, 3, 4] thiadiazole and imidazo [2, 1-b] thiazole," *Farmaco*, vol. 47, no. 1, pp. 63–75, 1992.
- [19] K. C. Joshi, P. Chand, and A. Dandia, "Studies in spiroheterocycles-part II-reactions of fluorine containing indole-2,3-diones with 1,2-phenyldiamines & 2,3-diaminopyridine in different media," *Indian Journal of Chemistry B*, vol. 23, pp. 743–745, 1984, Chemical Abstracts, vol. 101, 211055, 1984.
- [20] E. Palaska, G. Şahin, P. Kelicen, N. T. Durlu, and G. Altinok, "Synthesis and anti-inflammatory activity of 1-acylthiosemicarbazides, 1,3,4-oxadiazoles, 1,3,4-thiadiazoles and 1,2,4-triazole-3-thiones," *Farmaco*, vol. 57, no. 2, pp. 101–107, 2002.
- [21] H. Schiff, "Mittheilungen aus dem universitätslaboratorium in pisa: eineneue reihe organischer basen," *Justus Liebigs Annalen Der Chemie*, vol. 131, no. 1, pp. 118–119, 1864.
- [22] R. B. Moffett and N. Rabjohn, *Organic Synthesis*, John Wiley & Sons, Inc, New York, NY, USA, 1963.
- [23] J. H. Billman and K. M. Tai, "Reduction of Schiff bases. II. Benzhydrylamines and structurally related compounds," *The Journal of Organic Chemistry*, vol. 23, no. 4, pp. 535–539, 1958.
- [24] W. A. White and H. Weingarten, "A versatile new enamine synthesis," *Journal of Organic Chemistry*, vol. 32, no. 1, pp. 213–214, 1967.
- [25] F. Texier-Boullet, "A simple, convenient, and mild synthesis of imines on alumina surface without solvent," *Synthesis*, vol. 6-7, pp. 679–681, 1985.
- [26] H. Naeimi, F. Salimi, and K. Rabiei, "Mild and convenient one pot synthesis of Schiff bases in the presence of P₂O₅/Al₂O₃ as

- new catalyst under solvent-free conditions," *Journal of Molecular Catalysis A*, vol. 260, no. 1-2, pp. 100-104, 2006.
- [27] J. Zhu, L. Chen, H. Wu, and J. Yang, "Highly efficient procedure for the synthesis of Schiff bases using hydrotalcite-like materials as catalyst," *Chinese Journal of Chemistry*, vol. 27, no. 10, pp. 1868-1870, 2009.
- [28] A. Naqvi, M. Shahnawaaz, A. V. Rao, D. S. Seth, and N. K. Sharma, "Synthesis of schiff bases via environmentally benign and energy-efficient greener methodologies," *E-Journal of Chemistry*, vol. 6, no. 1, pp. S75-S78, 2009.
- [29] D. J.-N. Uppiah, M. G. Bhowon, and S. J. Laulloo, "Solventless synthesis of imines derived from diphenyldisulphide diamine or p-Vanillin," *E-Journal of Chemistry*, vol. 6, no. 1, pp. S195-S200, 2009.
- [30] M. Gopalakrishnan, P. Sureshkumar, V. Kanagarajan, and J. Thanusu, "New environmentally-friendly solvent-free synthesis of imines using calcium oxide under microwave irradiation," *Research on Chemical Intermediates*, vol. 33, no. 6, pp. 541-548, 2007.
- [31] A. M. Asiri, S. A. Khan, H. M. Marwani, and K. Sharma, "Synthesis, spectroscopic and physicochemical investigations of environmentally benign heterocyclic Schiff base derivatives as antibacterial agents on the bases of *in-vitro* and density functional theory," *Journal of Photochemistry and Photobiology B*, vol. 120, pp. 82-89, 2013.
- [32] S. Patil, S. D. Jadhav, and S. K. Shinde, "CES as an efficient natural catalyst for synthesis of Schiff bases under solvent-free conditions: an innovative green approach," *Organic Chemistry International*, vol. 2012, Article ID 153159, 5 pages, 2012.
- [33] H. Sachdeva, D. Dwivedi, and S. Khaturia, "Aqua mediated facile synthesis of 2-(5/7-fluorinated-2-oxoindolin-3-ylidene)-N-(4-substituted phenyl) hydrazine carbothioamides," *Research Journal of Pharmaceutical, Biological and Chemical Sciences*, vol. 2, no. 2, pp. 213-219, 2011.
- [34] H. Sachdeva and D. Dwivedi, "Lithium-acetate-mediated biginelli one-pot multicomponent synthesis under solvent-free conditions and cytotoxic activity against the human lung cancer cell line A549 and breast cancer cell line MCF7," *The Scientific World Journal*, vol. 2012, Article ID 109432, 9 pages, 2012.
- [35] A. Dandia, R. Singh, H. Sachdeva, and K. Arya, "Microwave assisted one pot synthesis of a series of trifluoromethyl substituted spiro [indole-triazoles]," *Journal of Fluorine Chemistry*, vol. 111, no. 1, pp. 61-67, 2001.
- [36] H. Sachdeva, R. Saroj, S. Khaturia, and D. Dwivedi, "Operationally simple green synthesis of some Schiff bases using grinding chemistry technique and evaluation of antimicrobial activities," *Green Processing and Synthesis*, vol. 1, no. 5, pp. 469-477, 2012.
- [37] S. Patil, S. D. Jadhav, and U. P. Patil, "Natural acid catalyzed synthesis of Schiff base under solvent-free condition: as a green approach," *Archives of Applied Science Research*, vol. 4, no. 2, pp. 1074-1078, 2012.
- [38] R. Breslow, "Hydrophobic effects on simple organic reactions in water," *Accounts of Chemical Research*, vol. 24, no. 6, pp. 159-164, 1991.
- [39] S. Otto and J. B. F. N. Engberts, "Hydrophobic interactions and chemical reactivity," *Organic and Biomolecular Chemistry*, vol. 1, no. 16, pp. 2809-2820, 2003.
- [40] J. Chandrasekhar, S. Shariffskul, and W. L. Jorgensen, "QM/MM simulations for Diels-Alder reactions in water: contribution of enhanced hydrogen bonding at the transition state to the solvent effect," *The Journal of Physical Chemistry B*, vol. 106, no. 33, pp. 8078-8085, 2002.
- [41] A. Lubineau and J. Auge, "Water as solvent in organic synthesis," *Topics in Current Chemistry*, vol. 206, pp. 1-39, 1999.
- [42] A. Libineau, J. Auge, and Y. Queneau, "Water-promoted organic reactions," *Synthesis*, no. 8, pp. 741-760, 1994.
- [43] H. Sachdeva, R. Saroj, S. Khaturia, and D. Dwivedi, "Environmentally friendly synthesis and characterization of some new 1, 2, 4-triazole derivatives as organic fluorescent materials and potent fungicidal agents," *Organic Chemistry International*, vol. 2013, Article ID 659107, 19 pages, 2013.
- [44] M. Curini, F. Epifano, F. Maltese, and M. C. Marcotullio, "Novel chiral Schiff base ligands from amino acid amides and salicylaldehyde," *Tetrahedron Letters*, vol. 43, no. 21, pp. 3821-3823, 2002.
- [45] N. Sari and P. Gürkan, "Some novel amino acid-schiff bases and their complexes synthesis, characterization, solid state conductivity behaviors and potentiometric studies," *Zeitschrift für Naturforschung B*, vol. 59, no. 6, pp. 692-698, 2004.
- [46] V. Kanagarajan, M. R. Ezhilarasi, and M. Gopalakrishnan, "One-pot' ultrasound irradiation promoted synthesis and spectral characterization of an array of novel 1,1'-(5,5'-(1,4-phenylene) bis(3-aryl-1H-pyrazole-5,1(4H,5H)-diyl))diethanones, a bis acetylated pyrazoles derivatives," *Spectrochimica Acta A*, vol. 78, no. 2, pp. 635-639, 2011.
- [47] A. K. Mansour, M. M. Eid, and N. S. A. M. Khalil, "Synthesis and reactions of some new heterocyclic carbohydrazides and related compounds as potential anticancer agents," *Molecules*, vol. 8, no. 10, pp. 744-755, 2003.
- [48] G. Kaur and R. Yadav, "Anti-cancer activities of various heterocyclic containing entites—a review," *International Journal of Natural Product Science*, vol. 1, article 104, 2012.
- [49] I. Ali, A. Haque, K. Saleem, and M. F. Hsieh, "Curcumin-I Knoevenagel's condensates and their Schiff's bases as anticancer agents: synthesis, pharmacological and simulation studies," *Bioorganic & Medicinal Chemistry*, vol. 21, no. 13, pp. 3808-3820, 2013.
- [50] H. Chen and J. Rhodes, "Schiff base forming drugs: mechanisms of immune potentiation and therapeutic potential," *Journal of Molecular Medicine*, vol. 74, no. 9, pp. 497-504, 1996.
- [51] V. Vichai and K. Kirtikara, "Sulforhodamine B colorimetric assay for cytotoxicity screening," *Nature Protocols*, vol. 1, no. 3, pp. 1112-1116, 2006.



Hindawi

Submit your manuscripts at
<http://www.hindawi.com>

