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An efficient synthesis of bis(indolyl)methanes and evaluation of their antimicrobial activities

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Abstract

A versatile and efficient method has been developed for the synthesis of bis(indolyl)methanes by using aluminium triflate (0.5 mol%) as a novel catalyst. Further, some of the synthesized compounds were evaluated for their efficacy as antibacterial and antifungal activities. Most of the compounds have shown moderate to good inhibitory activity.

Keywords: *Bis(indolyl)methanes, aluminium triflate, antibacterial activity, antifungal activity*

Introduction

Indole derivatives are found to exhibit a wide spectrum of pharmacological activities such as cytotoxic [1–2], antitumour [3], antiviral [3], antimicrobial [4] and anti-inflammatory activities [5]. Bis(indolyl)methanes (BIMs), which contain two indole or substituted indole units in a molecule, are found in bioactive metabolites of terrestrial and marine origin [6–8]. Over the past few years, much attention has been given for the search of specific bis(indole) secondary metabolites in view of their novel structural feature and broad spectrum of biological activities [9]. Recently, some bis(indole) alkaloids have been isolated from plants (I and II) and sponges (III and IV) wherein, two indole units are joined through different heterocyclic moieties. Further, such molecules have been also designed and synthesized that exhibit promising antibacterial, as well as antifungal activities ([10–13]; Figure 1).

Nitrofurans and isosteric nitrothiophenes are heterocyclic compounds that have been well documented as potential antimicrobial agents [14–18]. In spite of extensive research work that has been carried out in this class of compounds, there is still scope for the development of new molecules based on this type of heterocyclic systems [19–23]. It is well established that the activity of such nitroheterocycles is mainly due to the metabolic reduction of their nitro functionality by nitroreductase enzyme [24]. Moreover, a number of other furan derivatives have also exhibited antibacterial activity [25,26]. Therefore, in view of these findings, we have designed some new bisindoles by employing a new methodology. Furthermore, these compounds have been evaluated for their efficacy as antibacterial and antifungal agents.

It is well known in the literature that the reaction of indole with aromatic or aliphatic aldehydes and ketones produce azafulvenium salts, and this can undergo further addition with the second indole molecule to afford BIM [27]. Several other methods have been reported in the literature for the synthesis of BIMs using protic acid (e.g. HCl; [28–30]) or Lewis acids (e.g. $AlCl_3$, BF_3; [31,32]). These reactions have also been investigated using various triflates and chlorides [33–36]. More recently, Bhuyan and co-workers [37] have reported the synthesis of BIMs in protic solvents like methanol and water. However, this

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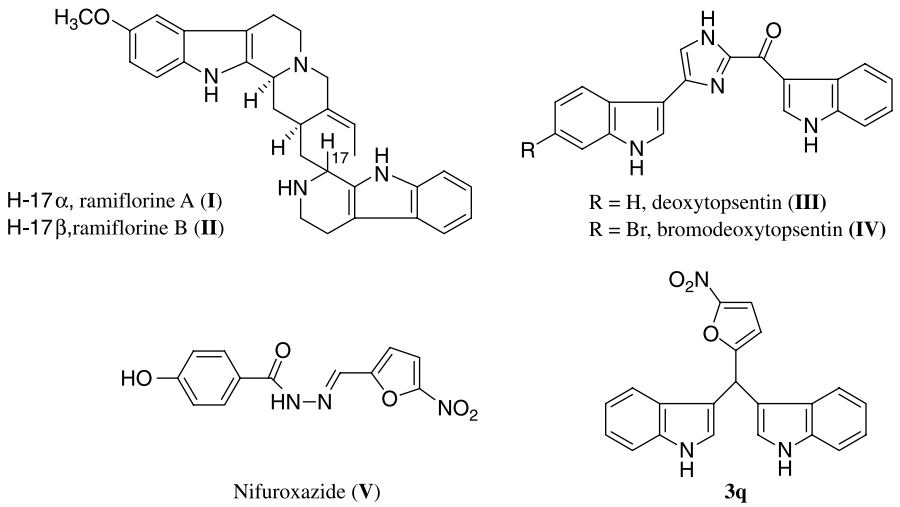


Figure 1. Chemical structures of bis(indole) alkaloids, **I, II** isolated from *Aspidosperma ramiflorum* and **III, IV** from sponge *Spongosorites* sp., nifuroxazide (**V**) and nitrofuryl BIM (**3q**).

method requires longer reaction time (1–20 h) and heterocyclic moieties like furfural 2-aldehyde, thiophene 2-aldehyde and aliphatic aldehydes did not react with indole in aqueous media, whereas in methanol low yields (16–65%) have been obtained. In today's context, if water can be replaced instead of organic solvents, it is significant from the environmental concerns point of view. However, this replacement is difficult for mainly two reasons; firstly, most of the organic substances are insoluble in water and secondly, many reactive substrates, reagents as well as catalysts are decomposed or deactivated by water. In recent years, aluminium triflouromethanesulfonate has been used for various transformations in organic synthesis [38–42]. Therefore, based on the versatility and usefulness of this catalyst, herein, we report a new and efficient method for the synthesis of BIMs catalyzed by 0.5 mol% $Al(OTf)_3$. Further, these compounds have been evaluated for their antimicrobial activity against various antibacterial and antifungal strains. Most of these compounds have shown good antibacterial and antifungal activities.

Materials and methods

Chemistry

^1H and ^{13}C NMRs were recorded on a Bruker UXNMR/XWIN-NMR (200 MHz) or Varian VXR-Unity (400 MHz) with TMS (0 ppm) as an internal standard. Coupling constants are reported in Hertz. EI mass spectra were recorded on a VG-7070H Micromass mass spectrometer at 200°C, 70 eV, with a trap current of 200 μA and 4 kV of acceleration voltage. LC–MS were recorded on instrument LC-MSD-Trap-SL and ESI MS on Micro mass, Quattro LC using ESI^+ software with capillary voltage 3.98 kV. Melting points were determined with an Electrothermal melting point apparatus and are reported uncorrected. IR spectra (KBr) were measured with a Thermo Nicolet Nexus 670 Spectrometer (ν_{max} in cm^{-1}). Analytical TLC of all reactions was performed on Merck prepared plates (silica gel 60 F-254 on glass). Column chromatography was performed using Acme silica gel. Starting materials were purchased from Sigma-Aldrich (Poole, UK). MeCN was distilled from CaH_2 and dried over 4 Å molecular sieves. Micro analytical data (C, H and N) agreed with the proposed structures within ±0.4% of the theoretical values. All the standard organisms were obtained from IMTECH Chandigarh, and nutrient agar and potato dextrose agar (PDA) were procured from Himedia Laboratories, Mumbai, India.

*General procedure for the synthesis of bis(indolyl) methanes (**3a–v**)*

To a mixture of indole (217 mg, 1 mmol), substituted aldehydes (0.5 mmol) in acetonitrile (5 ml), aluminium triflate (0.5 mol%) was added and stirred at room temperature for the appropriate time (Tables I and II). After completion of the reaction as monitored by TLC, water was added to the reaction mixture and products were extracted into ethyl acetate (3 × 20 ml). The combined organic layer was washed with brine, dried over anhydrous Na_2SO_4, concentrated in vacuum and purified by column chromatography, by using ethyl acetate–petroleum ether mixture as eluent to afford the desired products.

Spectral data for selected compounds

*3,3′-Bis-indolyl phenylmethane (**3a**)*. mp 148–150°C (lit.[17] 150–152°C); ^1H NMR (200 MHz, $CDCl_3$ + DMSO-d_6): δ 7.77 (bs, 2H, NH), 7.15–7.35

Table I. $Al(OTf)_3$ catalysed synthesis of BIM derivatives (**3a**–**v**) by the condensation of indole with various aldehydes.

Product	R	Time (min)	Yield (%)[a,b]
3a	C_6H_5	14	95
3b	2-Cl—C_6H_4	4	92
3c	4-Cl—C_6H_4	6	90
3d	4-Br—C_6H_4	30	80
3e	2-NO_2—C_6H_4	28	80
3f	3-NO_2—C_6H_4	30	80
3g	4-NO_2—C_6H_4	30	85
3h	4-Me—C_6H_4	20	95
3i	4-OMe—C_6H_4	12	92
3j	4-^{i}Pr—C_6H_4	3	95
3k	3,4,5-tri-OMe—C_6H_2	25	80
3l	3,4-di-Cl—C_6H_3	30	85
3m	2-OH—C_6H_4	55	75
3n	4-OH—C_6H_4	45	80
3o	3-OMe-4-OH—C_6H_3	35	75
3p	2-Furyl	14	80
3q	5-Nitrofuryl	35	82
3r	2-Thienyl	25	80
3s	5-Nitrothienyl	60	80
3t	CH_3	40	85
3u	C_2H_5	35	80
3v	C_4H_9	25	96

[a] Yield obtained with 0.5 mol% $Al(OTf)_3$; [b] Based on isolation by column chromatography and the products were characterized by NMR, IR and MS.

(m, 9H), 7.10 (t, $\mathcal{J} = 8.13$ Hz, 2H), 6.94 (t, $\mathcal{J} = 8.31$ Hz, 2H), 6.57 (d, $\mathcal{J} = 2.26$ Hz, 2H), 5.83 (s, 1H); ^{13}C NMR (50 MHz, $CDCl_3$ + DMSO-d_6): δ 136.0, 133.8, 130.1, 129.7, 129.7, 129.4, 126.8, 124.5, 122.6, 119.5, 119.0, 113.2, 39.3; LC-MS $m/z = 321.0$ $(M - 1)^+$, 345.1 $(M + Na)^+$; IR (KBr) (ν_{max}/cm^{-1}): 3395 (NH), 3050, 2924, 1596, 1455, 1088, 1004, 746. Anal. Calcd for $C_{23}H_{18}N_2$: C, 85.68; H, 5.63; N, 8.69. Found: C, 85.92; H, 5.54; N, 8.42.

3,3′-Bis-indolyl(4-nitrophenyl)methane (***3g***). mp 219–221 (lit.[17e] 217–219°C); ^{1}H NMR (200 MHz, $CDCl_3$ + DMSO-d_6): δ 10.50 (bs, 2H, NH), 8.15 (d, $\mathcal{J} = 8.56$ Hz, 2H), 7.57 (d, $\mathcal{J} = 8.56$ Hz, 2H), 7.39 (d, $\mathcal{J} = 7.81$ Hz, 2H), 7.28 (d, $\mathcal{J} = 7.81$ Hz, 2H), 7.09 (t, $\mathcal{J} = 7.81$ Hz, 2H), 6.91 (t, $\mathcal{J} = 7.81$ Hz, 2H), 6.73 (d, $\mathcal{J} = 2.34$ Hz, 2H), 6.00 (s, 1H); ^{13}C NMR (50 MHz, $CDCl_3$ + DMSO-d_6): δ 136.3, 134.2, 129.4, 129.0, 128.5, 124.1, 123.2, 122.4, 121.5, 118.4, 118.2, 112.5, 39.2; LC-MS $m/z = 366$ $(M - 1)^+$; IR (KBr) (ν_{max}/cm^{-1}): 3420 (NH), 3050, 2923, 1594, 1505, 1454, 1340, 1242, 1095, 1008, 743. Anal. Calcd for $C_{23}H_{17}N_3$: C, 75.19; H, 4.66; N, 11.44. Found: C, 75.44; H, 4.54; N, 11.68.

3,3′-Bis-indolyl(4-methoxyphenyl)methane (***3i***). mp 183–185°C (lit.[17e] 187–189°C); ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.88 (bs, 2H, NH), 7.35 (q, $\mathcal{J} = 7.55$ Hz, 4H), 7.23 (d, $\mathcal{J} = 8.30$ Hz, 2H), 7.15 (t, $\mathcal{J} = 7.55$ Hz, 2H), 6.98 (t, $\mathcal{J} = 7.55$ Hz, 2H), 6.80 (d, $\mathcal{J} = 8.30$ Hz, 2H), 6.62 (d, $\mathcal{J} = 2.26$ Hz, 2H), 5.82 (s, 1H), 3.78 (s, 3H, —OCH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 143.7, 136.2, 133.6, 130.8, 130.2, 128.7, 126.8, 124.5, 122.6, 120.2, 119.6, 114.8, 55.4, 39.6; LC-MS $m/z = 353.1$ $(M + 1)^+$, 375.0 $(M + Na)^+$; IR (Neat) (ν_{max}/cm^{-1}): 3394 (NH), 3054, 2925, 1608, 1507, 1554, 1336, 1241, 1090, 1025, 742. Anal. Calcd for $C_{24}H_{20}N_2O$: C, 81.79; H, 5.72; N, 7.95. Found: C, 81.92; H, 5.81; N, 7.60.

3,3′-Bis-indolyl furfurylmethane (***3p***). mp > 300°C (lit.[17] > 300°C); ^{1}H NMR (200 MHz, $CDCl_3$ + DMSO-d_6): δ 10.23 (bs, 2H, NH), 7.33 (q, $\mathcal{J} = 7.03$ Hz, 5H), 7.02 (t, $\mathcal{J} = 7.03$ Hz, 2H), 6.80–6.95 (m, 4H), 6.25 (q, $\mathcal{J} = 1.56$ Hz, 1H), 6.00 (d, $\mathcal{J} = 3.12$ Hz, 1H), 5.84 (s, 1H); ^{13}C NMR (50 MHz, $CDCl_3$ + DMSO-d_6): δ 157.0, 140.5, 136.1, 126.1, 122.8, 120.8, 118.8, 118.2, 115.8, 110.9, 109.6, 105.7, 33.5; ESI MS $m/z = 311.27$ $(M - 1)^+$; IR (KBr) (ν_{max}/cm^{-1}): 3406 (NH), 3052, 2924, 1614, 1453, 1336, 1091, 1006, 739. Anal. Calcd for $C_{21}H_{16}N_2O$: C, 80.95; H, 5.26; N, 8.97. Found: C, 80.61; H, 5.17; N, 9.19.

3,3′-Bis-indolyl(5-nitrofurfuryl)methane (***3q***). mp 97°C (charred); ^{1}H NMR (200 MHz, $CDCl_3$ + DMSO-d_6): δ 10.60 (bs, 2H, NH), 7.27–7.42 (m, 5H), 7.05 (t, $\mathcal{J} = 7.03$ Hz, 2H), 6.85–6.99 (m, 4H), 6.32 (d, $\mathcal{J} = 3.90$ Hz, 1H), 5.96 (s, 1H); ^{13}C NMR (50 MHz, $CDCl_3$): δ 161.5, 150.4, 136.0, 126.9, 125.4, 123.1, 120.7, 118.2, 118.0, 112.8, 110.9, 109.9, 33.9; LC-MS $m/z = 380$ $(M + Na)^+$; IR (KBr) (ν_{max}/cm^{-1}): 3407 (NH), 3053, 2926, 1721, 1485, 1354, 1239, 1094, 1015, 741. Anal. Calcd for $C_{21}H_{15}N_3O_3$: C, 70.58; H, 4.23; N, 11.76. Found: C, 70.32; H, 4.11; N, 11.79.

3,3′-Bis-indolyl thienylmethane (***3r***). mp 155–157°C (lit.[18] 150–153°C); ^{1}H NMR (200 MHz, $CDCl_3$ + DMSO-d_6): δ 10.21 (bs, 2H, NH), 7.35 (t, $\mathcal{J} = 8.57$ Hz, 4H), 7.10–7.15 (m, 1H), 7.05 (dt, $\mathcal{J} = 7.85$, 1.43 Hz, 2H), 6.85–6.95 (m, 6H), 6.10 (s, 1H); ^{13}C NMR (50 MHz, $CDCl_3$ + DMSO-d_6): δ 143.2, 132.0, 126.2, 121.6, 121.2, 119.8, 118.6, 117.5, 115.3, 114.5, 106.5, 33.6; ESI MS $m/z = 327.20$ $(M - 1)^+$; IR (KBr) (ν_{max}/cm^{-1}): 3403 (NH), 3077, 2925, 1616, 1454, 1333, 1213, 1086, 1002, 744. Anal. Calcd for $C_{21}H_{16}N_2S$: C, 76.80; H, 4.91; N, 8.53. Found: C, 76.56; H, 4.74; N, 8.76.

3,3′-Bis-indolyl(5-nitrothienyl)methane (***3s***). mp 181°C (charred); ^{1}H NMR (200 MHz, $CDCl_3$ + DMSO-d_6): δ 10.60 (bs, 2H, NH), 7.80 (d, $\mathcal{J} = 4.68$ Hz, 1H),

Table II. Antibacterial activity[a] of BIMs expressed in zone of inhibition in millimetres.

	Gram-positive				Gram-negative							
	B. Subtilis		*S. aureus*		*E. coli*		*P. aeruginosa*		*P. oleovorians*		*K. pneumoniae*	
Compound	100 μg/ml	150 μg/ml	100 μg/ml	150 μg/ml	100 μg/ml	150 μg/ml	100 μg/ml	150 μg/ml	100 μg/ml	150 μg/ml	100 μg/ml	150 μg/ml
3a	8	9	10	12	10	11	9	10	10	11	–	–
3 g	9	10	10	11	9	10	8	10	8	9	–	–
3i	9	10	–	9	11	12	–	9	9	10	–	–
3p	11	14	14	16	12	14	12	14	13	14	11	12
3q	16	17	11	13	14	17	12	14	17	18	12	14
3r	8	10	10	11	11	13	9	10	9	10	–	–
3s	10	12	11	12	11	14	10	11	12	13	10	10
3t	15	16	14	16	17	17	10	11	15	16	12	14
3u	16	18	14	17	15	18	15	19	16	18	11	15
3v	12	13	13	14	12	13	10	13	12	13	11	12
Strept	19		21		29		24		29		23	

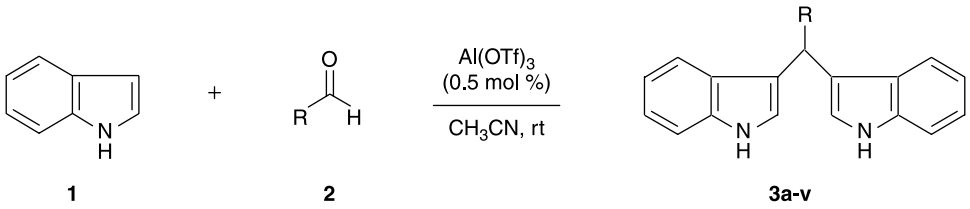
[a]The tests were performed in duplicate and repeated thrice; –, bacteria are resistant to the compound at the concentrations; Strept, Streptomycin (50 μg/ml).

7.39 (d, $\mathcal{J} = 8.59$ Hz, 4H), 7.10 (t, $\mathcal{J} = 7.81$ Hz, 2H), 6.90–7.02 (m, 5H), 6.14 (s, 1H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 15.9, 148.1, 135.7, 127.9, 125.1, 123.9, 122.7, 120.5, 117.9, 117.8, 115.4, 110.7, 35.2; LC-MS $m/z = 396.9$ $(M + Na)^+$; IR (KBr) (ν_{max}/cm^{-1}): 3444 (NH), 3056, 2922, 1525, 1458, 1335, 1212, 1090, 737. Anal. Calcd for $C_{21}H_{15}N_3SO_2$: C, 67.54; H, 4.05; N, 11.25. Found: C, 67.28; H, 4.12; N, 11.38.

*3,3′-Bis-indolyl ethane (**3t**)*. mp 153–155°C (lit.[17] 148–150°C); ^{1}H NMR (200 MHz, $CDCl_3$ + DMSO-d_6): δ 9.95 (bs, 2H, NH), 7.45 (d, $\mathcal{J} = 8.01$ Hz, 2H), 7.28 (d, $\mathcal{J} = 8.01$ Hz, 2H), 7.00 (t, $\mathcal{J} = 8.01$ Hz, 2H), 6.91 (d, $\mathcal{J} = 2.18$ Hz, 2H), 6.85 (d, $\mathcal{J} = 7.28$ Hz, 2H), 4.60 (q, $\mathcal{J} = 6.55$ Hz, 1H), 1.77 (d, $\mathcal{J} = 6.55$ Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 135.7, 125.5, 120.3, 119.7, 119.4, 118.0, 116.9, 110.2, 27.0, 20.0; ESI MS $m/z = 259.20$ $(M - 1)^+$; IR (KBr) (ν_{max}/cm^{-1}): 3413 (NH), 3075, 2957, 1622, 1455, 1336, 1094, 1011, 737. Anal. Calcd for $C_{18}H_{16}N_2$: C, 83.05; H, 6.19; N, 10.76. Found: C, 83.40; H, 6.30; N, 10.62.

*3,3′-Bis-indolyl propane (**3u**)*. Semi solid; ^{1}H NMR (200 MHz, $CDCl_3$ + DMSO-d_6): δ 10.00 (bs, 2H, NH), 7.46 (d, $\mathcal{J} = 7.81$ Hz, 2H), 7.27 (d, $\mathcal{J} = 7.81$ Hz, 2H), 6.92–7.06 (m, 4H), 6.86 (t, $\mathcal{J} = 7.03$ Hz, 2H), 4.29 (t, $\mathcal{J} = 7.03$ Hz, 1H), 2.20 (p, $\mathcal{J} = 7.03$ Hz, 2H), 0.98 (t, $\mathcal{J} = 7.03$ Hz, 3H); ^{13}C NMR (50 MHz, $CDCl_3$ + DMSO-d_6): δ 135.9, 127.4, 122.3, 120.6, 120.5, 116.0, 115.2, 109.3, 30.6, 24.8, 8.2; LC-MS $m/z = 273.1$ $(M - 1)^+$; IR (KBr) (ν_{max}/cm^{-1}): 3414 (NH), 3053, 2926, 2865, 1615, 1456, 1340, 1093, 1012, 744. Anal. Calcd for $C_{19}H_{18}N_2$: C, 83.18; H, 6.61; N, 10.21. Found: C, 83.01; H, 6.80; N, 10.33.

*3,3′-Bis-indolyl pentane (**3v**)*. Semi solid; ^{1}H NMR (200 MHz, $CDCl_3$ + DMSO-d_6): δ 7.60 (bs, 2H, NH), 7.52 (d, $\mathcal{J} = 7.55$ Hz, 2H), 7.16–7.23 (m, 2H), 7.07 (dt, $\mathcal{J} = 6.79$, 1.51 Hz, 2H), 6.97 (dt, $\mathcal{J} = 6.79$, 1.51 Hz, 2H), 6.0 (dd, $\mathcal{J} = 2.66$, 0.75 Hz, 2H), 4.40 (t, $\mathcal{J} = 7.55$ Hz, 1H), 2.17 (q, $\mathcal{J} = 7.55$ Hz, 2H), 1.20–1.40 (m, 4H), 0.87 (t, $\mathcal{J} = 6.79$ Hz, 3H); ^{13}C NMR (50 MHz, $CDCl_3$ + DMSO-d_6): δ 136.8, 126.2, 123.1, 119.7, 119.8, 117.0, 116.0, 109.5, 34.2, 31.5, 26.7, 22.2, 12.2; LC-MS $m/z = 301.1$ $(M - 1)^+$; IR (KBr) (ν_{max}/cm^{-1}): 3413 (NH), 3054, 2927, 2858, 1617, 1456, 1339, 1094, 1011, 742. Anal. Calcd for $C_{21}H_{22}N_2$: C, 83.40; H, 7.33; N, 9.26. Found: C, 83.16; H, 7.42; N, 9.39.



Scheme 1. Synthesis of BIMs.

Antibacterial assay

The antibacterial activity of the synthesized compounds was determined by the well diffusion method according to Linday [43]. Three to five identical colonies from each agar plate were lifted with a sterile wire loop and transferred into a tube containing 5 ml of nutrient agar. The turbidity of each bacterial suspension was adjusted to reach an optical comparison to that of a 0.5 McFarland standard; resulting in a suspension containing approximately $1-2 \times 10^8$ CFU/ml. Nutrient agar plates were inoculated by streaking the swab over the entire sterile agar surface. This procedure was repeated by streaking two more times, rotating the plate approximately 60° each time to ensure uniform distribution of the inoculum. As a final step, the rim of the agar was also swabbed. After allowing the inoculum to dry at room temperature, 6 mm diameter wells were prepared in the agar with the help of sterilized cork borer. The different concentrations of the test compounds (100–150 μg/ml) were prepared by dissolving in dimethyl sulfoxide (DMSO) and introduced into duplicate wells. The plates were incubated at 37°C for 24 h. Subsequently, the plates were examined for bacterial growth inhibition and the inhibition zone diameter measured to the nearest millimeter. The standard antibiotic (streptomycin 50 μg/well) was used as positive control, whereas, the equivalent amount of solvent (DMSO) did not exhibit any activity in the assay.

Antifungal assay

The method followed for antifungal bioassay is similar to that followed for antibacterial assay, where the medium is PDA 39 g/l. All the test compounds were studied for their antifungal activity at concentration 100–150 μg/ml using DMSO as a solvent. The solvent did not exhibit any activity at the concentrations used. The treated and the controls were kept in an incubator at 28 ± 2°C for 48 h and inhibition zones were measured to the nearest millimeter. Three replicates were maintained for each treatment. Amphotericin-B (50 μg/ml) was used as positive control.

Results and discussion

Chemistry

BIMs were obtained by the condensation of indole (**1**) and various aldehydes (**2a**–**v**) in acetonitirle at room temperature in presence of 0.5 mol% $Al(OTf)_3$ in high yields in shorter reaction time (Scheme 1, Table I). Aromatic aldehydes with electron deficient substituents required longer reaction times to produce comparable yields than those of their simple and electron-rich counterparts. Electron rich aldehydes like anisaldehyde, reacted rapidly using indole within 12 min (compound **3i**), whereas, aliphatic aldehydes gave good yields of their corresponding products in 25–40 min (compounds **3t**–**v**). To best of our knowledge, this is the first report for the synthesis of BIMs using this catalyst which possess more advantages over earlier methods like mild reaction conditions, cleaner reactions, shorter reaction times, high yield of products and low catalytic amount.

Antibacterial activity

Amongst the synthesized compounds **3a**, **3g**, **3i** and **3p**–**v** were screened for their antibacterial activity against *Escherichia coli* (MTCC 443), *Pseudomonas aeruginosa* (MTCC 1688), *Pseudomonas oleovorans* (MTCC 617), *Klebsiella pneumoniae* (MTCC 618) as Gram-negative bacteria, *Bacillus subtilis* (MTCC 441), *Staphylococcus aureus* (MTCC 96), as Gram-positive bacteria and the antifungal activity was evaluated against yeast *Candida albicans* (MTCC 227) and filamentous fungi *Rhizopus oryzae* (MTCC-262), *Aspergillus niger* (MTCC 1344). The inhibitory zones (in mm) were determined by using agar well method (cup plate method). Antibiotics, streptomycin and amphotericin-B were used as positive controls against bacteria and fungi, respectively. In all determinations, tests were performed in duplicate and results were reported as mean of at least three determinations. Table II showed that all the tested compounds exhibited moderated to good antibacterial activity. Compounds **3q**, **3t** and **3u** showed significant inhibition against all the bacteria tested and were not strain dependent. In compounds **3a**, **3g** and **3i**, there was no effect of activating or deactivating substituents on the aryl ring in their inhibitory activity and found to be less potent than the other compounds tested. Moreover, these compounds were inactive against *K. pneumoniae* at all the concentrations used. The incorporation of 5-nitro furfuroyl group in BIM (**3q**) enhanced the activity in all the strains except *P. aeruginosa* than the corresponding furfuroyl derivative

Table III. Antifungal activity of BIMs expressed in zone of inhibition in millimetres.

	Yeast		Filamentous fungi			
	C. albicans		*R. oryzae*		*A. niger*	
Compd	100 μg/ml	150 μg/ml	100 μg/ml	150 μg/ml	100 μg/ml	150 μg/ml
3a	10	12	–	–	–	–
3g	10	14	–	–	–	–
3i	–	–	–	–	–	–
3p	–	–	10	12	–	–
3q	8	9	8	9	10	11
3r	10	12	–	–	–	–
3s	9	10	8	9	9	11
3t	12	14	18	20	12	14
3u	10	13	13	15	14	18
3v	–	–	10	12	–	–
Amph	23		23		22	

–, No activity has shown; Amph, Amphotericin B (50 μg/ml).

(**3p**) and activity reduced against *S. aureus*. Similarly, 5-nitrothiophenyl derivative (**3s**) was found to be more potent than the corresponding thiophenyl derivative (**3r**). The significant inhibition exhibited by the compounds **3q** and **3s** might be due to the presence of nitro group on the fifth position of their corresponding hetero aryl rings. Among the heteroaryl methyl(bisindoles) the activity is in the order **3q** > **3s** > **3p** > **3r**. The replacement of aryl group with alkyl chain on the 3-methyl of bisindoles (**3t**–**v**) also enhanced the significant inhibitory activity. Compound **3u** was found to be the most active from this series against all the bacterial strains tested.

Antifungal activity

The investigation of antifungal screening data from Table III revealed that all the tested compound showed moderate to good fungal inhibition. Compounds **3q** and **3s**–**u** were active against all the fungal species. The 5-nitro heteroaryl derivatives (**3q** and **3s**) exhibited better inhibitory activity against all the three fungal strains than the corresponding heteroaryl derivatives, whereas, compound **3p** was mild active only against *R. oryzae* and **3r** active only against *C. albicans*. Compound **3t** was found to be the most active against *C. albicans* and *R. oryzae* and **3u** was the most active against *A. niger*.

Conclusions

In conclusion, we have developed an efficient synthetic protocol for the synthesis of BIMs from various aldehydes and indoles using 0.5 mol% $Al(OTf)_3$. Furthermore, the synthesized compounds using this method have been evaluated for their antibacterial and antifungal activities. Almost all the compounds showed moderate to good antibacterial and antifungal activities. 5-Nitro heteroaryl containing BIM (**3q** and **3s**) have exhibited significant antibacterial activity. Compound **3u** was found to be the most active from the tested compound in this series. Compounds **3s**–**u** demonstrated significant antifungal activity. Further research in this area is in progress in our laboratory.

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References

[1] Sakemi S, Sun HH. Nortopsentins A, B, and C. Cytotoxic and antifungal imidazolediyl bis[indoles] from the sponge *Spongosorites ruetzleri*. J Org Chem 1991;56:4304–4307.

[2] Bao B, Sun Q, Yao X, Hong J, Lee CO, Sim CJ, Im KS, Jung JH. Cytotoxic bisindole alkaloids from a marine sponge *Spongosorites* sp. J Nat Prod 2005;68:711–715.

[3] Wright AE, Pomponi SA, Ctross SS, McCarthy PJ. A new bis-(indole) alkaloid from a deep-water marine sponge of the genus *Spongosorites*. J Org Chem 1992;57:4772–4775.

[4] Gunasekera SP, McCarthy PJ, Kelly-Borges M. Hamacanthins A and B, new antifungal bis indole alkaloids from the deep-water marine sponge, *Hamacantha* Sp. J Nat Prod 1994;57: 1437–1441.

[5] Mancini I, Guella G, Pietra F, Debitus C, Waikedre J. From inactive nortopsentin D, a novel bis(indole) alkaloid isolated from the axinellid sponge *Dragmacidon* sp. from deep waters south of new caledonia, to a strongly cytotoxic derivative. Helv Chim Acta 1996;79:2075–2082.

[6] Fahy E, Potts BCM, Faulkner DJ, Smith K. 6-Bromotryptamine derivatives from the Gulf of California tunicate *Didemnum candidum*. J Nat Prod 1991;54:564–569.

[7] Bell R, Carmeli S, Sar N. Vibrindole A, a metabolite of the marine bacterium, *Vibrio parahaemolyticus*, isolated from the toxic mucus of the boxfish *Ostracion cubicus*. J Nat Prod 1994; 57:1587–1590.

[8] Garbe TR, Kobayashi M, Shimizu N, Takesue N, Ozawa M, Yukawa H. Indolyl carboxylic acids by condensation of indoles with α-keto acids. J Nat Prod 2000;63:596–598.

[9] Faulkner DJ. Marine natural products. Nat Prod Rep 2002;19: 1–49.

[10] Tanaka JCA, da Silva CC, de Oliveria AJB, Nakamura CV, Filho BPD. Antibacterial activity of indole alkaloids from *Aspidosperma ramiflorum*. Braz J Med Bio Res 2006;39: 387–391.

[11] Oh K-B, Mar W, Kim S, Kim J-Y, Lee T-H, Kim J-G, Shin D, Sim CJ, Shin J. Antimicrobial activity and cytotoxicity of bis(indole) alkaloids from the sponge *Spongosorites* sp. Biol Pharm Bull 2006;29:570–573.

[12] Prudhomme M, Sancelme M, Bonnefoy A, Fabbro D, Meyer T. Synthesis and antimicrobial activities of mono-indolyl- and bisindolyloximes. Eur J Med Chem 1999;34: 161–165.

[13] Tiwari RK, Singh D, Singh J, Yadav V, Pathak AK, Dabur R, Chhillar AK, Singh R, Sharma GL, Chandra R, Verma AK. Synthesis and antibacterial activity of substituted 1,2,3,4-tetrahydropyrazino[1,2-*a*]indoles. Bioorg Med Chem Lett 2006;16:413–416.

[14] Szarvasi E, Fontaine L, Betbeder-Matibet A. Antimicrobials. New nitrofuran derivatives. J Med Chem 1973;16:281–287.

[15] Hoyle W, Roberts GP, Meth-Cohn O. Potential antimicrobial furans. J Med Chem 1973;16:709–710.

[16] Islip PJ, Johnson MR. Nitrofuryl heterocyclics. 3. J Med Chem 1973;16:1308–1310.

[17] Burch HA, Benjamin LE, Russell HE, Freedman R. Nitrofurfuryl heterocycles. 12. 4-Amino-6-(5-nitro-2-furyl)isoxazolo[5,4-*d*]pyrimidines and 4-amino-2-(5-nitro-2-furyl) pyrimido[4,5-*d*]pyrimidines. J Med Chem 1974;17:451–453.

[18] Wang CY, Chiu CW, Muraoka K, Michie PD, Bryan GT. Antibacterial activity of nitropyrroles, nitrothiophenes, and aminothiophenes *in vitro*. Antimicrob Agents Chemother 1975;8:216–219.

[19] Masunari A, Tavares LC. A new class of nifuroxazide analogues: Synthesis of 5-nitrothiophene derivatives with antimicrobial activity against multidrug-resistant *Staphylococcus aureus*. Bioorg Med Chem 2007;15:4229–4236.

[20] Foroumadi A, Mansouri S, Kiani Z, Rahmani A. Synthesis and *in vitro* antibacterial evaluation of *N*-[5-(5-nitro-2-thienyl)-1,3,4-thiadiazole-2-yl] piperazinyl quinolones. Eur J Med Chem 2003;38:851–854.

[21] Pires JR, Saito C, Gomes SL, Giesbrecht AM, Amaral AT. Investigation of 5-nitrofuran derivatives: Synthesis, antibacterial activity, and quantitative structure–activity relationships. J Med Chem 2001;44:3673–3681.

[22] Monasterios M, Escorche M, Avendano M. Conformational analysis, electronic properties and molecular electrostatic potential of nitrofurans derivatives with antibacterial activity. J Mol Structure 2005;748:49–55.

[23] Holla BS, Akberali PM, Shivananda MK. Studies on nitrophenylfuran derivatives: Part XII. Synthesis, characterization, antibacterial and antiviral activities of some nitrophenylfurfurylidene-1,2,4-triazolo[3,4-*b*]-1,3,4-thiadiazines. II Farmaco 2001;56:919–927.

[24] Kedderis GL, Miwa GT. The metabolic activation of nitroheterocyclic therapeutic agents. Drug Metab Rev 1988; 19:33–62.

[25] Holla BS, Rao BS, Shridhara K, Akberali PM. Studies on arylfuran derivatives: Part XI. Synthesis, characterisation and biological studies on some mannich bases carrying 2,4-dichlorophenylfurfural moiety. II Farmaco 2000;55:338–344.

[26] Holla BS, Akberali PM, Shivananda MK. Studies on arylfuran derivatives: Part X. Synthesis and antibacterial properties of arylfuryl-Δ^2-pyrazolines. II Farmaco 2000;55:256–263.

[27] Yadav JS, Reddy BVS, Murthy CVSR, Kumar GM, Madan Ch. Lithium perchlorate catalyzed reactions of indoles: An expeditious synthesis of bis(indolyl)methanes. Synthesis 2001;5:783–787.

[28] Gregorovich BV, Liang K, Clugston M, Macdonald S. Reductive C-alkylation. Can J Chem 1968;46:3291–3300.

[29] Roomi M, Macdonald S. Cyclizative condensation. I. 2-methylindole with acetone and methyl ethyl ketone. Can J Chem 1970;48:139–143.

[30] Auria M. Photochemical synthesis of diindolylmethanes. Tetrahedron 1991;47:9225–9230.

[31] Chatterjee A, Manna S, Benerji J, Pascard C, Prange T, Shoolery J. Lewis-acid-induced electrophilic substitution in indoles with acetone. Part 2. J Chem Soc Perkin Trans 1980;1: 553–555.

[32] Noland WE, Venkiteswaran MR, Richards CG. Cyclizative condensations. I. 2-Methylindole with acetone and methyl ethyl ketone. J Org Chem 1961;26:4241–4248.

[33] Chen DP, Yu LB, Wang PG. Lewis acid-catalyzed reactions in protic media. Lanthanide-catalyzed reactions of indoles with aldehydes or ketones. Tetrahedron Lett 1996;37:4467–4470.

[34] Nagarajan R, Perumal PT. $InCl_3$ and $In(OTf)_3$ catalyzed reactions: Synthesis of 3-acetyl indoles, bis-indolylmethane and indolylquinoline derivatives. Tetrahedron 2002;58:1229–1232.

[35] Mi XL, Luo SZ, He JQ, Chen JP. $Dy(OTf)_3$ in ionic liquid: An efficient catalytic system for reactions of indole with aldehydes/ketones or imines. Tetrahedron Lett 2004;45: 4567–4570.

[36] Ji SJ, Wang SY, Zhang Y, Loh TT. Facile synthesis of bis(indolyl)methanes using catalytic amount of iodine at room temperature under solvent-free conditions. Tetrahedron 2004; 60:2051–2055.

[37] Bhuyan JP, Deb ML. An efficient and clean synthesis of bis(indolyl)methanes in a protic solvent at room temperature. Tetrahedron Lett 2006;47:1441–1443.

[38] Olah GA, Farooq O, Farnia SMF, Olah JA. Friedel–Crafts chemistry. 11. Boron, aluminum, and gallium tris(trifluoromethanesulfonate) (triflate): Effective new Friedel–Crafts catalysts. J Am Chem Soc 1988;110:2560–2565.

[39] Kamal A, Khan MNA, Reddy KS, Srikanth YVV, Krishnaji T. $Al(OTf)_3$ as a highly efficient catalyst for the rapid acetylation of alcohols, phenols and thiophenols under solvent-free conditions. Tetrahedron Lett 2007;48:3813–3818.

[40] Firouzabadi H, Iranpoor N, Kohmareh G. Aluminium trifluromethanesulfonate [$Al(OTf)_3$] as a highly efficient and chemoselective catalyst for thioacetalization of carbonyl compounds under mild conditions. Synth Commun 2003; 33:167–173.

[41] Williams DBG, Lawton M. Aluminium triflate: A remarkable Lewis acid catalyst for the ring opening of epoxides by alcohols. Org Biomol Chem 2005;3:3269–3272.

[42] Williams DBG, Lawton M. Aluminium triflate: An efficient recyclable Lewis acid catalyst for the aminolysis of epoxides. Tetrahedron Lett 2006;47:6557–6560.

[43] Linday ME. Practical introduction to microbiology. London: E & F.N. Spon Ltd; 1962. p 177.