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Research Article

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Design and Synthesis of Novel Substituted (R) 4-[2-Pyridylmethoxy] Phenyl Methyl Sulphinyl Substituted Benzimidazoles and Evaluation of their Biological Activity

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ABSTRACT

A series of substituted (R)-4-[2-pyridylmethoxy] phenyl methyl sulphinyl substituted benzimidazole derivatives 10a to 10l were synthesized by asymmetric oxidation of 4-[2-pyridylmethoxy] phenyl methyl thio substituted benzimidazoles (7a to 7l). All the targeted compounds were characterized by IR, ^{1}H NMR and Mass spectral studies. The anti-microbial activity of synthesized compounds were screened against different strains of bacteria (S.Aureus, E. Fecalis, S. Mutans, Klebsiella) with reference to the standard ciprofloxacin and fungi (A.Niger, A.Flavus) was evaluated with reference to the standard Fluconazole. The results showed that some of the synthesized (10a-10l) compounds exhibited moderate anti-bacterial and anti-fungal activities, among which compounds 10g, 10h and 10i exhibited about 36% zone of inhibition activities against E. Fecalis, Klebsiella and A.Niger. Further these compounds were screened for anti-inflammatory activities, these synthesized title compounds exhibited as potent against MMP-2 and MMP-9.

Keywords: Asymmetric synthesis; Sulphoxide; Anti-inflammatory; Anti-fungal; Anti-bacterial

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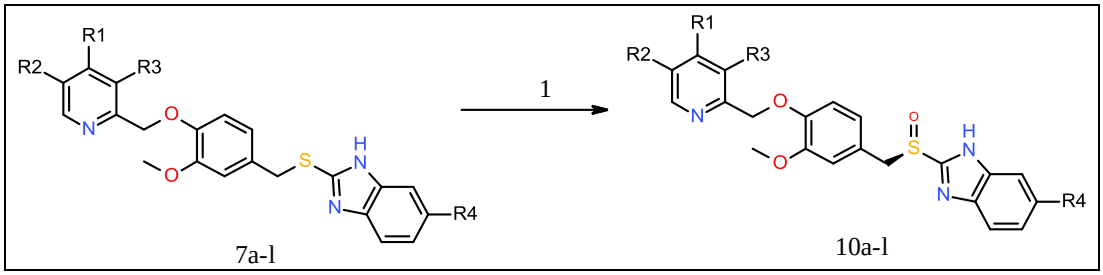
INTRODUCTION

Anti-microbial research over for the last five decades has been focused to meet medical needs to treat medical infectious disease caused by life threatening pathogens. The introduction of variety of anti-microbial agents in multiple unconnected drug classes, resistance continues to emerge. Due to the essential requirement of pharmaceutical research and to support them, anti-microbial research was taken over by the academic research to face the clinical challenges by bringing forward new agents with anti-microbial activity against microbes. Due to the rapid development of bacterial resistance to anti-microbial agents, it was vital to discover novel scaffold for the drug substance designs and synthesis of new microbial agents to help in the battle against pathogenic microorganisms. Benzimidazole is a hetero cyclic organic compound with bicyclic ring systems, which is a core structure in various synthetic pharmaceuticals in medicinal chemistry, enhances a broad spectrum of biological activities including anti-bacterial [1,2] and anti-fungal [3] activities. Several benzimidazole derivatives have been reported to show significant activities such as anti-microbial [4], anti-inflammatory [5], anti-hypertensive [6], anti-viral [7], analgesic [8], anti-histamine [9], anti-convulsant [10], anti-ulcer [11], anti-cancer [12], anti-oxidant [13]. Optically activity was first described in 1926, this discovery was helpful for discussions of the sulphur-oxygen bond in sulphoxide, [S-O] and non-polarity of sulphur. Later, chiral sulphoxides are slowly emerged as a class of compounds in asymmetric synthesis. Further the 'Methyl sulphoxide' compounds, which are important group in active pharmaceutical ingredients in drug industries, and these have variety of biological activities. Compared to racemic sulphoxide, enantiopure sulphoxides are more important chiral compounds in asymmetric synthesis, and some also have useful biological activities, such as

anti-ulcer (Proton Pump Inhibitors) [14], anti-bacterial, anti-fungal, anti-atherosclerotic [15], as well as anti-hypertensive and anti-proliferative [16]. Since the definite demand for synthesis of novel sulphoxide compounds is increasing, and there exists a tremendous scope for development.

In our previous work, a serious of 4-[2-pyridylmethoxy] phenyl methyl thio substituted benzimidazole derivatives were synthesised and reported their biological activities got tested against S.Aureus, E. Fecalis, S. Mutans, Klebsiella and A.Niger, A.Flavus and Candida [17]. Based on the above details available in literature, it was thought that worthwhile to synthesise some new compounds by asymmetric oxidation of 4-[2-pyridylmethoxy] phenyl methyl thio substituted benzimidazole derivatives to study their biological activities. As an extension of this topic, we have designed and synthesized 12 novel substituted (R) 4-[2-pyridylmethoxy] phenyl methyl sulphinyl substituted benzimidazole derivatives as described in scheme 1 and table 1.

Scheme 1: Synthesis of substituted (R)-4-[2-pyridylmethoxy] phenyl methyl sulphinyl substituted benzimidazoles derivatives (10a to 10l)



Reagents: 1) Toluene, Diethyl D-tartrate, Titanium isopropoxide, Diisopropylethyl amine, Cumene hydroperoxide

Table 1: Synthesis of substituted (R) 4-[2-pyridylmethoxy] phenyl methyl sulphonyl substituted benzimidazoles derivatives (10a to 10l)

Comp	% of Yield	(+) SOR	R1	R2	R3	R4
10a	72	27.6	$-OCH_2CF_3$	-H	-CH3	-H
10b	75	32.38	$-OCH_2CF_3$	-H	-CH3	$-OCH_3$
10c	77	34.12	$-OCH_2CF_3$	-H	-CH3	$-OCHF_2$
10d	79	27.54	$-OCH_3$	$-CH_3$	-CH3	-H
10e	73	30.12	$-OCH_3$	$-CH_3$	-CH3	$-OCH_3$
10f	74	25.54	$-OCH_3$	$-CH_3$	-CH3	$-OCHF_2$
10g	45	31.38	$-OCH_3$	-H	$-OCH_3$	-H
10h	72	40.28	$-OCH_3$	-H	$-OCH_3$	$-OCH_3$
10i	47	35.34	$-OCH_3$	-H	$-OCH_3$	$-OCHF_2$
10j	74	28.8	$-O(CH_2)_3OCH_3$	-H	-CH3	-H
10k	68	18.62	$-O(CH_2)_3OCH_3$	-H	-CH3	$-OCH_3$
10l	71	8.16	$-O(CH_2)_3OCH_3$	-H	-CH3	$-CHF_2$

EXPERIMENTAL SECTION

The majority of the solvents were obtained from Sigma-Aldrich. The progress was monitored by thin-layer chromatography (TLC) using pre-coated SiO_2 gel (HF_{254} 200 mesh) aluminium plates (E Merck). Visualization was achieved with UV lights. Melting points of synthesized compounds were found out using Buche apparatus with one end opened capillary tube and all melting points were uncorrected. Specific optical rotation of these compounds were recorded on AUTOPOL-V analyser. The Infra-Red (IR) spectra of these compounds were recorded on ABB Bomen FTIR spectrometer MB 104 with KBr Pellets. ^{1}H NMR and Spectra were recorded on 400 MHz- Broker DPX 200 spectrometer by using tetramethyl silane (TMS) as an internal standard. Chemical shifts were reported as in parts per million (ppm) down fields from tetramethyl silane. Spin multiplicities were described as s (singlet), bs (broad singlet), d (doublet), t (triplet), q (quartet), p (pentet) and m (multiplet). Coupling constants were reported in Hertz (Hz).Mass spectra were recorded on Finnigan MAT 8230 mass spectrometer at 200°C, 70 eV with trap current of 200 IA, and 4 kV acceleration voltages.

Key starting materials for the preparation of substituted 4-[2-pyridylmethoxy] phenyl methyl sulfinyl and enantioselective sulphoxides were obtained from substituted 4-[2-pyridylmethoxy] phenyl methylthio substituted benzimidazole (7a-l) which was prepared using a procedures as reported in our earlier communication [16], and raw materials like Diethyl L-tartrate, Diethyl D-tartrate, Titanium isopropoxide, diisopropylethyl amine, Hydrogen peroxide, ammonium molybdate were as obtained from commercially available sources like Sigma-Aldrich and SDF chemicals.

General procedure for preparation of (R)-2-(4-((4-(2, 2, 2-trifluoroethoxy)-3-methylpyridin-2-yl) methoxy)-3-methoxybenzylthio)-1H-benzo[d] imidazole, 10a-l: Substituted 4-[2-pyridylmethoxy] phenyl methylthio-substituted-benzimidazole (**7a-l**) 2.0 gm (0.004 moles) was charged into anhydrous methylene dichloride and to this was charged with 0.84 gm of diethyl L-tartrate (0.004 moles), titanium isopropoxide (0.002 moles), and diisopropylethyl amine (0.004 moles), with catalytic quantity of water treated with a solution of Cumene hydroperoxide (0.0048 moles), heated to reflux maintained for 5 to 10 minutes, cooled to 0±5ºC stirred for 2 to 4 hours. After the TLC complies, the reaction mass was quenched with aqueous sodium metabisulphite solution, layer separated, aqueous layer was extracted with MDC. MDC layer was distilled and isolated with n-Heptane and dried under vacuum, and purified by using silica column to obtain title compounds 10a-l.

(R)-2-(4-((4-(2,2,2-Trifluoroethoxy)-3-methylpyridin-2-yl)methoxy)-3-methoxybenzylsulfinyl)-1Hbenzo[d] imidazole (10a): Off-white colour solid with 72% of yield; $C_{24}H_{22}F_3N_3O_4S$: MR 120-123°C, IR (KBr, ν_{max} in cm^{-1}): 3428 (N-H), 3075 (C-H, Aromatic ring), 2893 (C-H, Methoxy), 1 (C=N, Imidazole),1584 (C=N, Aromatic ring), 1515 (C=C, Aromatic ring), 1167 (C-N, Imidazole),1133 (C-O-C), 1039 (C-F), 752 (C-S), ^{1}H NMR (DMSO-d6) δ: 2.165 (s, 3H), 3.300 (s, 3H), 4.368-4.399 (d, 1H), 4.585-4.617 (d, 1H), 5.090 (s, 2H), 5.149 (s, 2H), 6.473 (s, 1H), 6.699-6.716 (d, 1H), 6.989-7.009 (d, 1H), 7.127-7.147 (d, 1H), 7.247-7.281 (m, 2H), 7.625 (s, 2H), 8.317-8.330 (d, 1H), 13.2 (bs, 1H), MS m/z 505.9$[M+H]^+$

(R)-2-(4-((4-(2,2,2-Trifluoroethoxy)-3-methylpyridin-2-yl)methoxy)-3-methoxybenzylsulfinyl)-5-methoxy-1H-benzo[d]imidazole (10b): Light cream colour solid with 75% of yield; $C_{25}H_{24}F_3N_3O_5S$: MR 98.3-96.8°C, IR (KBr, ν_{max} in cm^{-1}): 3424 (N-H), 3075 (C-H, Ar), 2962 (C-H, Methoxy), 1626 (C=N, Imidazole),1585 (C=N, Aromatic ring), 1516 (C=C, Aromatic ring), 1166 (C-N, Imidazole),1135 (C-O-C), 1026 (C-F), 773 (C-S), ^{1}H NMR (DMSO-d6) δ: 1.217 (s, 3H), 2.182 (s, 3H), 3.797 (s, 3H), 4.377-4.408 (d, 1H), 4.577-4.609 (d, 1H), 4.902-4.920 (d, 2H), 5.105 (s, 2H), 6.522 (s, 1H), 6.522-7.142 (m, 5H), 7.525-7.618 (d, 1H), 8.337 (s, 1H) , 13.244 (s, 1H), MS m/z 535.8 $[M+H]^+$

(R)-2-(4-((4-(2,2,2-Trifluoroethoxy)-3-methylpyridin-2-yl)methoxy)-3-methoxybenzylsulfinyl) 5 (difluoro methoxy)-1H-benzo[d]imidazole (10c): Light cream colour solid with 77% of Yield; $C_{25}H_{22}F_5N_3O_5S$: MR 97-98°C, IR (KBr, ν_{max} in cm^{-1}): 3415 (N-H), 2963 (C-H, Ar), 1627 (C=N, Imidazole),1584 (C=N, Aromatic ring), 1516 (C=C, Ar), 1167 (C-N, Imidazole),1135 (C-O-C), 1040 (C-F), 773 (C-S), ^{1}H NMR (DMSO-d6) δ: 2.175 (s, 3H), 3.340 (s, 3H), 4.367-4.397 (d, 1H), 4.590-4.620 (d, 1H), 4.902-4.917 (d, 2H), 5.098 (s, 2H), 6.711, 7.134 and 7.336 (t, 1H, Due to C-F Coupling), 6.496-6.616 (dd, 2H), 7.011-7.419 (m, 5H), 7.644-7.660 (d, 1H), 8.329 (s, 1H), MS m/z 572.2 $[M+H]^+$

(R)-2-(4-((4-Methoxy-3, 5-dimethylpyridin-2-yl) methoxy)-3-methoxybenzylsulfinyl)-1H-benzo[d]imidazole (10d): White colour solid with 79% of Yield; $C_{24}H_{25}N_3O_4S$: MR 136-138°C, IR (KBr, ν_{max} in cm^{-1}): 3416 (N-H), 3064 (C-H, Ar), 2934 (C-H, Methoxy), 1589 (C=N, Imidazole),1605 (C=N, Aromatic ring), 1516 (C=C, Ar), 1156 (C-N, Imidazole),1108 (C-O-C), 1003 (C-F), 733 (C-S), ^{1}H NMR(DMSO-d6) δ: 2.215 (s, 6H), 3.331 (s, 3H), 3.727 (s, 3H), 4.378-4.411 (d, 1H), 4.594-4.626 (d, 1H), 5.049 (s, 2H), 6.467 (s, 1H), 6.703-6.721 (d, 1H), 7.006-7.026 (d, 1H), 7.296 (broad s, 2H), 7.531-7.733 (m, 2H), 8.200 (s, 1H), 13.354 (s, 1H) , MS m/z 452.2 $[M+H]^+$

(R)-2-(4-((4-Methoxy-3,5-dimethylpyridin-2-yl)methoxy)-3-methoxybenzylsulfinyl)-5-methoxy-1H benzo [d]imidazole (10e): White colour solid with 73% of Yield; $C_{25}H_{27}N_3O_5S$: MR 172-173°C, IR (KBr, ν_{max} in cm^{-1}): 3419 (N-H), 3079 (C-H, Ar), 2832 (C-H, Methoxy), 1625 (C=N, Imidazole),1589 (C=N, Ar), 1515 (C=C, Aromatic ring), 1150 (C-N, Imidazole),1105 (C-O-C), 775 (C-S), ^{1}H NMR (DMSO-d6) δ: 2.214 (s, 6H), 3.356 (s, 3H), 3.722 (s, 3H), 3.787 (s, 3H), 4.362-4.393 (s, 1H), 4.567-4.599 (s, 1H), 5.046 (s, 2H), 6.517 (s, 1H), 6.703-6.723 (d, 1H), 6.881-6.886 (d, 1H), 6.903-6.908 (d, 1H), 7.003-7.024 (d, 1H), 7.066 (s, 1H), 7.514-7.536 (d, 1H), 8.195 (s, 1H), MS m/z, 482.3 $[M+H]^+$

(R)-2-(4-((4-Methoxy-3, 5-dimethylpyridin-2-yl) methoxy)-3-methoxybenzylsulfinyl)-5-(difluoromethoxy)-1H-benzo[d]imidazole (10f): White colour solid with 72% of Yield; $C_{25}H_{25}F_2N_3O_5S$: MR 168-169°C, IR (KBr, ν_{max} in cm^{-1}): 3413 (N-H), 3068 (C-H, Ar), 2939 (C-H, Methoxy), 1631 (C=N, Imidazole),1590 (C=N, Ar), 1516 (C=C, Ar), 1160 (C-N, Imidazole),1133 (C-O-C), 1032 (C-F), 776 (C-S), ^{1}H NMR (DMSO-d6) δ: 2.215 (s, 6H), 3.324 (s, 3H), 3.726 (s, 3H), 4.366-4.398 (d, 1H), 4.592-4.623 (s, 1H), 4.592-4.623 (d, 1H), 5.048 (s, 2H), 6.471 (s, 1H), 6.753, 7.027 and 7.275 (t, 1H, Due to C-F coupling), 6.685-7.565 (m, 4H), 7.776-7.796 (d, 1H), 8.196 (s, 1H), 13.483 (s, 1H), MS m/z, 518.2 $[M+H]^+$

(R)-2-(4-((3,4-Dimethoxypyridin-2-yl)methoxy)-3-methoxybenzylsulfinyl)-1H-benzo[d]imidazole(10g): Off white colour solid with 45% of Yield; $C_{23}H_{23}N_3O_5S$: MR 151-152°C, IR (KBr, ν_{max} in cm^{-1}): 3067 (C-H, Ar), 2943 (C-H, Methoxy), 1735 (C=N, Imidazole),1585 (C=N, Ar), 1511 (C=C, Ar), 1266 (C-N, Imidazole), 1146 (C-O-C), 747 (C-S), ^{1}H NMR (DMSO-d6) δ: 3.265 (s, 3H), 3.734 (s, 3H), 3.891 (s, 3H), 4.382-4.413 (d, 1H), 4.594-4.625 (d, 1H), 4.995 (s, 2H), 6.438 (s, 1H), 6.712-6.729 (d, 1H), 7.006-7.026 (d, 1H), 7.129-7.141 (d, 1H), 7.290-7.742 (m, 4H), 8.197-8.209 (d, 1H), 13.339 (s, 1H), MS m/z 454.2 $[M+H]^+$

(R)-2-(4-((3,4-Dimethoxypyridin-2-yl)methoxy)-3-methoxybenzylsulfinyl)-5-methoxy-1H-benzo[d]imidazole (10h): Light orange colour solid with 72% of Yield; $C_{24}H_{25}N_3O_6S$: MR 137-138°C, IR (KBr, ν_{max} in cm^{-1}): 3424 (N-H), 3045 (C-H, Ar), 2835 (C-H, Methoxy), 1625 (C=N, Imidazole),1588 (C=N, Ar), 1512 (C=C, Ar), 1148 (C-N, Imidazole),1114 (C-O-C), 722 (C-S), ^{1}H NMR (DMSO-d6) δ: 3.625 (s, 3H), 3.739 (s, 3H), 3.794 (s, 3H), 3.893 (s, 3H), 4.378-4.408 (d, 1H), 4.569-4.601 (d, 1H), 4.997 (s, 2H), 6.487 (s, 1H), 6.710-6.728 (d, 1H), 6.891-7.633 (m, 5H), 8.197-8.210 (d, 1H), 13.214 (s, 1H), MS m/z 484.2 $[M+H]^+$

(R)-2-(4-((3,4-Dimethoxypyridin-2-yl)methoxy)-3-methoxybenzylsulfinyl)-5-(difluoromethoxy)-1H benzo [d]imidazole (10i): White colour solid with 47% of Yield; $C_{24}H_{23}F_2N_3O_6S$: MR 126-127°C, IR (KBr, ν_{max} in cm^{-1}): 3440 (N-H), 3942 (C-H, Ar), 2841 (C-H, Methoxy), 1625 (C=N, Imidazole),1589 (C=N, Aromatic ring), 1513 (C=C, Ar), 1164 (C-N, Imidazole),1124 (C-O-C), 1034 (C-F), 722 (C-S), ^{1}H NMR (DMSO-d6) δ: 3.288 (s, 3H), 3.630 (s, 3H), 3.889 (s, 3H), 4.375-4.407 (d, 1H), 4.598-4.630 (d, 1H), 5.000 (s, 2H), 7.053, 7.426 and 7.786 (t, 1H, Due to C-F Coupling), 6.439 (s, 1H), 6.700-6.719 (d, 1H), 7.009-7.029 (d, 1H), 7.123-7.786 (m, 3H), 8.195-8.208 (d, 1H), 13.492 (s, 1H), MS m/z 520.2 $[M+H]^+$

(R)-2-(4-((4-(3-Methoxypropoxy)-3-methylpyridin-2-yl)methoxy)-3-methoxybenzylsulfinyl)-1H-benzo[d] imidazole (10j): White colour solid with 74% of Yield; $C_{26}H_{29}N_3O_5S$: MR 114-115°C, IR (KBr, ν_{max} in cm^{-1}): 3433 (N-H), 3065 (C-H, Ar), 2829 (C-H, Methoxy), 1584 (C=N, Ar), 1517 (C=C, Ar), 1190 (C-N, Imidazole),1152 (C-O-C), 747 (C-S), ^{1}H NMR (DMSO-d6) δ: 1.964-1.994 (t, 2H), 2.137 (s, 3H), 3.245 (s, 3H), 3.306-3.333 (d, 3H), 3.470-3.500 (t, 2H), 4.091-4.120 (t, 2H), 4.373-4.406 (d, 1H), 4.585-4.617 (d, 1H), 5.061 (s, 2H), 6.472 (s, 1H), 6.697-6.717 (d, 1H), 6.996-7.016 (d, 2H), 7.293 (s, 2H), 7.514 (s, 1H), 7.737 (s, 1H), 8.204-8.254 (d, 1H), 13.327 (s, 1H), MS m/z 496.1 $[M+H]^+$

(R)-2-(4-((4-(3-Methoxypropoxy)-3-methylpyridin-2-yl)methoxy)-3-methoxybenzyl sulfinyl)-5-methoxy-1H-benzo[d]imidazole (10k): Off white colour solid with 68% of Yield; $C_{27}H_{31}N_3O_6S$: MR 123-124°C, IR (KBr, ν_{max} in cm^{-1}): 3406 (N-H), 3075 (C-H, Aromatic ring), 2883 (C-H, Methoxy), 1625 (C=N, Imidazole),1586 (C=N, Aromatic ring), 1512 (C=C, Aromatic ring), 1148 (C-N, Imidazole),1125 (C-O-C), 731 (C-S), ^{1}H NMR (DMSO-d6) δ: 1.967-1.996 (p, 2H), 2.140 (s, 3H), 2.500 (s, 3H), 3.245 (s, 3H), 3.332 (s, 3H), 3.472-3.500 (t, 2H), 4.110 (s, 2H), 4.367-4.399 (d, 1H), 4.590-4.623 (d, 1H), 5.068 (s, 2H), 6.481 (s, 1H), 6.687-6.705 (d, 1H), 6.999-7.012 (d, 2H), 7.147 (s, 1H), 7.236 (s, 1H), 7.776 (s, 1H), 8.249-8.262 (d, 1H), 13.478 (s, 1H), MS m/z 525.9 $[M+H]^+$

(R)-2-(4-((4-(3-Methoxypropoxy)-3-methylpyridin-2-yl) methoxy)-3-methoxybenzyl sulfinyl)-5-(difluoro methoxy)-1H-benzo[d]imidazole (10l): Light brown colour solid with 71% of Yield; $C_{27}H_{29}F_2N_3O_6S$: MR 115-116°C, IR (KBr, ν_{max} in cm^{-1}): 3426 (N-H), 3119 (C-H, Ar), 2834 (C-H, Methoxy), 1626 (C=N, Imidazole), 1586 (C=N, Ar), 1514 (C=C, Ar), 1166 (C-N, Imidazole),1127 (C-O-C), 1039 (C-F), 774 (C-S), 1H NMR (DMSO-d6) δ: 1.912 (t/s, 2H), 1.960-1.988 (t, 3H), 3.580-3.608 (d, 3H), 3.671-3.690 (d, 2H), 3.791 (s, 3H), 4.102 (s, 2H), 4.370-4.401 (d, 1H), 4.571-4.603 (d, 1H), 5.069 (s, 2H), 6.511 (s, 1H), 6.701-6.719 (d, 1H), 6.925 (s, 1H), 6.997-7.007 (m, 4H), 7.606 (s, 1H), 8.253-8.266 (d, 1H), 13.247 (s, 1H). MS m/z 562.0 $[M+H]^{+\cdot}$

Anti-bacterial activity

The anti-fungal activity studies of the novel synthesized compounds (10a-l) were screened using disk diffusion method and results were tabulated in table 2. The medium used for disk diffusion procedure was Brain Heart Infusion agar. The medium was cooled to room temperature. The inoculums were prepared using a loop or swab and transferred the colonies to the plates. Turbidity was adjusted with broth to equal that of a 0.5 McFarland turbidity standard that had vortexes within 15 minutes. Those were alternatively, standardized the suspension with a photometric device. The sterile cotton swab was dipped into the inoculums and it was rotated against the wall of the tube above the liquid to remove excess inoculums. Swab entire surface of agar plate was rotated three times approximately 60º between streaking to ensure even distribution. The aerosols were avoided by not hitting the sides of the petriplate. The inoculated plate was allowed to stand for atleast 3 minutes but no longer than 15 min before making wells. The compound was added into the plate by using 5mm diameter hollow tube

and heated then a pressed the inoculated, the agar plate was removed immediately by making a well in the plate, which was made five well on each plate. Then the compound was added with concentration of 100, 125 µl and 150 µl into the respective wells on each plate. The plate was incubated within 15 minutes. The incubated plate was monitored for 18-24 hours at 37°C in CO2 jar, confluent or nearly confluent plates was taken for reading and measured the diameter of inhibition zone to nearest millimeter by holding measuring device.

Evaluation of the anti-bacterial of these synthesized compounds 10a-l were screened by using pathogenic organisms of three gram positive (S.Aureus, S.Mutans and E. Fecalis) and gram negative (Klebsiella) by measuring the inhibition zone in mm at 100, 125 and 150 µg/ml concentration, Ciprofloxacin was used as standard and was also screened under similar conditions.

Table 2: Zone of inhibition of anti-bacterial activity of synthesized compounds (10a to 10l)

Comp	*S.Aureus*			*E. Fecalis*			*S.Mutans*			*Klebsiella*		
	[a]100	[a]150	[a]125	[a]100	[a]150	[a]125	[a]100	[a]150	[a]125	[a]100	[a]150	[a]125
10a	15	13	10	16	13	13	10	R	R	18	15	10
10b	R	R	R	18	15	12	20	15	10	20	18	15
10c	18	15	R	20	18	15	R	R	R	23	18	15
10d	R	R	R	25	23	20	R	R	12	20	18	13
10e	R	R	R	18	17	25	R	R	R	18	15	13
10f	15	13	12	25	24	20	R	R	R	23	20	18
10g	R	R	R	28	26	20	R	R	R	28	25	23
10h	R	R	R	25	23	21	R	R	10	18	15	13
10i	R	R	R	28	24	20	R	R	10	20	18	15
10j	R	R	R	20	13	R	R	R	8	23	20	18
10k	R	R	R	13	10	R	R	R	10	28	25	20
10l	13	12	10	18	15	10	R	R	9	20	18	15

[a] Concentration is given in micro gram/ mill litter, R is resistant, Standard is Ciprofloxacin

Anti-fungal activity

The anti-fungal activity studies of the novel synthesized compound (10a-l) were screened using disk diffusion method. The medium used for disk diffusion procedure was Sabouraud agar, was as explained in the above anti-bacterial study.

The evaluation of the anti-fungal of these synthesized compounds 10a-l were screened against three anti-fungal activities against A.Niger, A.Flavus and Candida by measuring the inhibition zone in mm at 50, 75, 100, 125 and 150 µg/ml concentration, Fluconazole was used as standard and also it was screened under similar conditions for comparison. The results of the anti-bacterial studies were summarized in table 3.

Table 3: Zone of inhibition of anti-fungal activity of synthesized compounds (10a to 10l)

Comp	*A.Niger*					*A.Flavus*				
	[a]150	[a]125	[a]100	[a]75	[a]50	[a]150	[a]125	[a]100	[a]75	[a]50
10a	33	30	25	23	20	R	R	R	R	R
10b	28	25	20	18	15	25	10	R	R	R
10c	18	15	13	10	8	15	12	9	R	R
10d	20	18	15	13	10	10	R	R	R	R
10e	18	16	15	12	10	15	10	R	R	R
10f	28	26	21	20	18	12	9	R	R	R
10g	32	30	29	26	25	14	8	R	R	R
10h	35	32	30	27	18	16	10	R	R	R
10i	36	33	31	28	25	12	8	R	R	R
10j	28	25	23	20	18	16	8	R	R	R
10k	18	15	13	10	8	18	14	10	R	R
10l	30	25	23	20	18	15	10	R	R	R

[a] Concentration is given in micro gram/ mill litter, R is resistant, Standard is Fluconazole

Anti-inflammatory

The evaluation of the anti-inflammatory of these synthesized compounds 10a-l were screened against MMP-2, and MMP-9 using Gelatin zymography method by measuring the Anti-inflammatory activity, Positive control (Tetracycline hydrochloride), Negative control (Tonsil tissue) sample was used as standard and also it was screened under similar conditions for comparison. The results of the anti-inflammatory studies were summarized in table 4.

Table 4: Anti-inflammatory activity of synthesized compounds (10a to 10l)

Comp	Activity against MMP-2	Activity against MMP-9
10a	80%	70%
10b	85%	65%
10c	90%	68%
10d	75%	50%
10e	60%	50%
10f	85%	45%
10g	55%	65%
10h	80%	70%
10i	90%	30%
10j	75%	40%
10k	80%	75%
10l	65%	85%

MMP-9 is Matrix Metallo Proteinase Type 9, MMP-2 is Matrix Metallo Proteinase Type 2, Positive control: 100%, Negative control: 10%, Positive control: Tetracycline hydrochloride, Negative control: Tonsil (tissue) sample

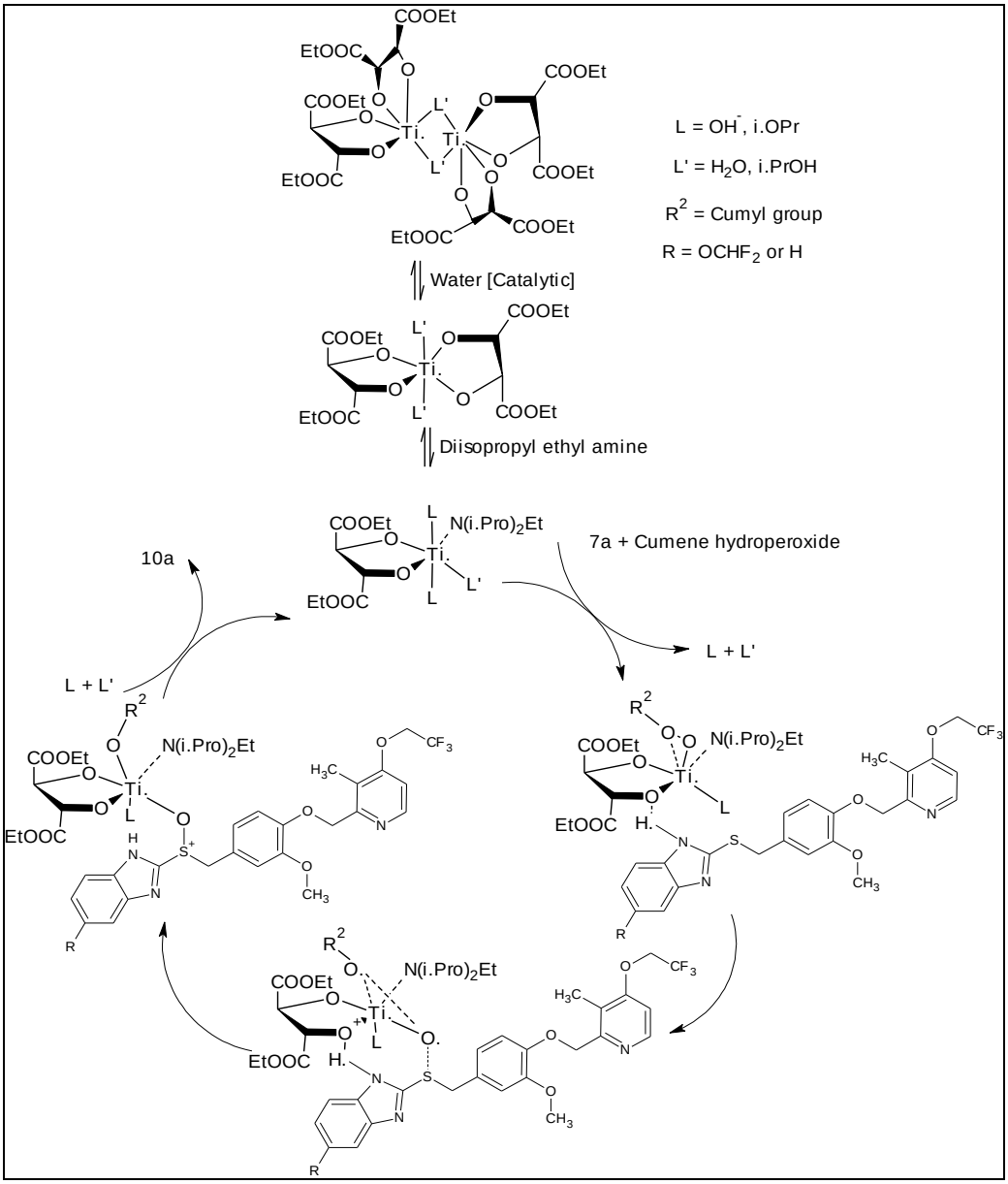
RESULTS AND DISCUSSION

The asymmetric sulfoxidation reaction of benzimidazole-based prochiral sulphides was studied to explore the mechanistic details of the highly efficient process (Figure 1), which is one of the industrial scale catalytic asymmetric procedures. The synthetic studies revealed that the smallest subunit governing the selectivity in the asymmetric oxidation process is an imidazole ring. Thus, by using the novel compound procedure methyl imidazole sulphide could be oxidized as efficiently as it is several functionalized derivatives including pyramidazole. However alkylation of the imidazole nitrogen led to a major drop of the enantioselectivity. Muthu Seenivasaperumal et al., reported that atmospheric pressure chemical ionisation mass spectrometry studies indicate that addition of small amount of water to the reaction mixture in presence of the diisopropylethyl amine, facilitates the formation of mononuclear titanium species which are the active catalytic intermediates of the enantio-selective oxidation reaction. Because of this formation of such mononuclear titanium species, chiral purity is more and yield of the required sulphoxide quantitative.

One of the most important features of the targeted procedures is that the diisopropylethyl amine addictive increase the enantioselectivity of the oxidation process. Under catalytic conditions, the amines are able to coordinate to titanium and dissociate the coordinated imidazole substrate. It was found that the oxidation requires a lower activation energy if the imidazole sulphides precursors does not coordinate to titanium. The presence of catalytic quantity of water helps to form mono titanium complex with substrate benzimidazole sulphide as depicted in the mechanism. The most important interaction governing the enantio selection is hydrogen bonding between the NH of the imidazole ring and the chiral tartrate ligand on titanium. When the oxidation reagent comes to contact with the complex, one oxygen of the dioxirane ring interacts with sulphur atom for the oxidation reaction which imposes an important structural constraint to the Transition State structure involving a linear arrangement of the peroxide oxygen and the sulphur atom. It is also observed that the formation of such mononuclear titanium complex, is facilitated by heating the benzimidazole sulphide compound with titanium isopropoxide, diethyl L-tartrate, diisopropylethyl amine and a catalytic quantity of water at ambient temperatures in chloroalkanes or aromatic hydrocarbons. If the water quantity is stoichiometric or more, Based on the above mechanistic aspects, we have arrived at a scalable process for the preparation of title compounds by using enantioselective oxidation of 4-[2-pyridylmethoxy] phenyl methylthio substituted benzimidazole (7a-l). Substituted 4-[2-pyridylmethoxy] phenyl methylthio substituted benzimidazole (7a-l) was charged into anhydrous methylene dichloride and this was charged with diethyl L-tartrate, titanium isopropoxide, and diisopropylethyl amine, with catalytic quantity of water and then heated to reflux, and maintained for 5 to 10 minutes. This resulting solution was cooled to 0±5ºC, added with a solution of Cumene hydroperoxide at 0 to 5˚C, stirred for 2 to 4 hours. After the TLC complies, the reaction mass was quenched with aqueous sodium metabisulphite solution, layer separated, and isolated title compounds 10a-l. The compounds were characterized by means of IR, 1H NMR, and MASS analysis.

From the anti-bacterial screening it was observed that some of the compounds exhibited anti-bacterial activity against all the organisms, these was tableted in table 2. The compounds 10a to 10l have showed highest activity against both gram positive (E. Fecalis) and gram negative (Klebsilla) bacteria. The compounds 10k showed maximum zone of inhibition (28) against Klebsilla. Only the compound 10b showed anti-bacterial activity against S. Mutans with three concentration levels. 10a, 10f and 10l which have only shown activity against S.Aureus. But all the synthesised compounds shown highest activity compare to the Ciprofloxacin standards drug with respect to E. Fecalis and Klebsilla.

Figure 1: Mechanism of an enantioselective oxidation of 7a by Cumene hydroperoxide using Titanium isopropoxide, Diethyl L tartrate and Cumene hydroperoxide



Further to the above study, the anti-fungal screening, was observed that all the compound exhibited highest anti-fungal activity against A. Niger fungi, which is tabulated in table 3. The compounds 10g, 10h and 10i showed maximum zone of inhibition (36) against A.Niger with all concentrations, which are containing two methoxy group in R2, R3 positions. These two electron rich groups [methoxy group] are increasing the electron density at the sulphoxide group when compared to the other substituents. The enhanced electron density may presumably be the reason behind the enhanced anti-fungal activity. All the compounds 10a-10l showed anti-fungal activity against A. Flavus which are only having concentration 150 µg/ml. In lower concentration these compounds have not shown any such activity. But all the synthesised compounds have shown higher anti-fungal activity when compared to the anti-bacterial activity. Further to the above study, the anti-inflammatory screening it was observed that all the compounds exhibited anti-inflammatory activity against the positive and negative controls which are as indicated in table 4. The compounds 10c and 10i have showed highest activity against MMP-2 which shown maximum activity 90% against reference tetracycline hydrochloride. The compound 10l which have shown maximum activity against MMP-9 with 85% positive control. Whatever groups present in R, R2, R3 and R4. But all the synthesised compound have shown highest activity compared to the anti-bacterial and anti-fungal activity.

CONCLUSION

The proposed synthetic scheme was found to be an efficient enantioselective method for the asymmetric oxidation of sulphides (7a-l) to corresponding R-sulphoxides (10a-l) at 0 to 5˚C. This enantioselective oxidation system is environmental safe, clean and with simple unit operations with quantitative yields. These novel compounds biologically active substituted benzimidazole sulphoxide compounds 10a-l possessing anti-inflammatory, anti-bacterial and anti-fungal activities. The anti-bacterial activity of these compounds was evaluated against gram positive and gram negative bacteria. The targeted compounds showed moderate anti-bacterial and anti-fungal activity compared to Ciprofloxacin and Fluconazole as standard reference drugs. All these title compounds predominantly show positive control on anti-inflammatory activity against MMP-2 & MMP-9.

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REFERENCES

[1] E Mentese; H Bektas; S Ulker; O Bekircan; B Kahveci. *J Enzyme Inhib Med Chem*, **2014**, 29, 64-68.
[2] R Janupally; VU Jeankumar; KA Bobesh; V Soni; PB Devi; VK Pulla; P Suryadevara; KS Chennubhotla; P Kulkarni; P Yogeeswari. *Bioorg Med Chem*, **2014**, 22, 5970-5987.
[3] H Goker; R Ertan; H Akgun. *Arch Der Pharm*, 199, 324(5), 283-291.
[4] N Sanahanbi; T Sivakumar. *Int J Pharm Bio Arc*, **2013**, 4(4), 717-722.
[5] S Gupta; SR Gupta; N Upmanyu; G Garg. *J Drug Des Med Chem*, **2015**, 1(2), 1216.
[6] JATR Kumar; JATL Jawahar; DP Pathak. *J Chem*, **2006**, 3(4), 278-285.
[7] AK Tiwari; A Mishra. Indian. *J Chem*, **2006**, 45B, 489-493.
[8] M Saraswathi; N Nagaraju; K Mnikanta; SK Mogalabi; C Eswaraiah; D Bardalai. *J Chem Pharm Res*, **2012**, 4(8), 3832-3836.
[9] T Nagase; T Mizutani; S Ishikawa; E Sekino; T Sasaki; T Fujimura. *J Med Chem*, **2008**, 1(15), 4780-4789.
[10] N Siddiqui; MD Shamsher Alam; R Ali; O Alam. *Med Chem Res*, **2016**, 25(7), 1390-1402.
[11] A Patil; S Ganguly; S Surana. Rasayan. *J Chem*, **2008**, 1(3), 447-460.
[12] SM Rida; SA El-Hawash; HT Fahmy; AA Hazzaa; MM El-Meligy. *Arch Pharm Res*, **2006**, 29(10), 826-833.
[13] KK Sumayya; VK Kamalabhai Amma; G Babu; CR Biju. *Int J Chem Pharm Sci*, **2013**, 1(3), 193-198.
[14] KC Lai; SK Lam; KM Chu; BCY Wong; WM Hui. *N Eng J Med*, **2002**, 346(26), 2033-2038.
[15] M Sovova; P Sova. Ceska. Slov. *Farm*, 2003, 52, 82-87.
[16] R Rajesh; N Nagaraju. *J Chem Pharm Res*, **2016**, 8(9), 228-237.
[17] TG Samir; OHE Ahmed; H Amer; MA Mamdouh; EEM Abeer; HH Andreas. *Sci Pharm*, **2016**, 84(1), 1-18.