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RESEARCH ARTICLE

SYNTHESIS AND BIOLOGICAL EVALUATION OF NOVEL IMIDAZOLE BASED COMPOUNDS

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Abstract

Objective: Some new imidazole derivatives (3i-xii) were synthesized as per design synthetic protocol scheme. The structures of newly prepared compounds were confirmed by modern analytical technique and elemental analysis.
Methods: All the synthetic compounds were screened for their antimicrobial activity against bacterial results showed good to remarkable activity. The MIC (minimum inhibitory concentration) values were determined by comparison to ciprofloxacin (anti-bacterial) and fluconazole (anti-fungal) as standard drug. Among them, compound 3iv and 3x exhibited notable antimicrobial activity. These compounds may be used as new template for the searching of potential antimicrobial agents.
Results: The purity of the compound was verified with the help of TLC (B: A, 9:1). % age of yield was found 83% and melting point noted 151-152°C. Compounds (3ii, 3viii and 3ix) were shown moderate activity against *E. coli*, *S. aureus*, *M. luteus* and *K. pneumonia*, whereas compounds (3iii, 3vii, 3xi and 3xii) showed mild activity against few bacterial strainsμg/ml. The compounds of electron releasing imidazole derivatives (3ii, 3iv, 3viii, 3ix, 3x and xi) presented comparatively better anti-fungal activity than the compounds of electron withdrawing imidazole derivatives (3iii, 3vii and 3xii).
Conclusion: The biological activity result revealed that all the newly synthetic compounds 3i-xii [4-(biphenyl-4-yl)-2-(substituted phenyl)-1H-imidazole] exhibited better antibacterial activity as compared to antifungal activity in compare to reference drug.
Keywords: Antibacterial activity, antifungal activity, biphenyl ethanone, imidazole.

INTRODUCTION

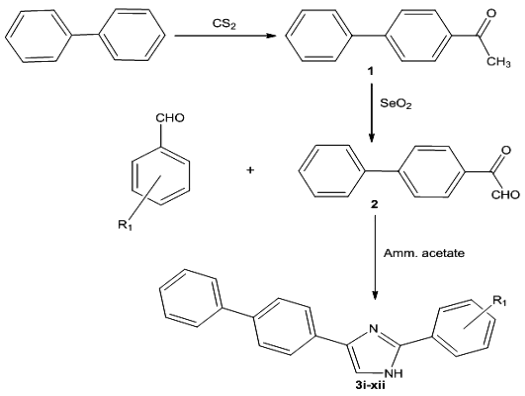
Nitrogen containing heterocyclic play vital role in pharmaceutical industries. The main aim of medicinal chemist in the recent times has been to develop drugs with enhanced their efficacy and duration of action and by decreasing their toxicities and side effects as well as creating new drugs by molecular modification[1,2]. The organic medicinal substances can be of natural or synthetic origin. The synthetic drugs are prepared by modifications of the structures of natural drugs, or by pure synthesis[3,4]. Over the years, innovations in new drug therapy has become, more complex, time consuming, costly, and the practicing medicinal chemists have been bombarded with surplus new methods and technologies to make the job of drug discovery more efficient[5]. Imidazoles are a class of five membered heterocyclic compounds having two nitrogen and three carbon atoms. Imidazole is an important group of compounds reported to have different biological activities and the present study was undertaken in order to synthesize some new derivatives of imidazole and related fused heterocyclic compounds and screen for their antimicrobial activity[6-11]. Among the various classes of heterocyclic compounds, imidazole is an important component of pharmacologically active compounds and a part of various available marketed drugs like Azathioprine (Leukemia), Metronidazole (Protozoal and antimicrobial activity, trichomoniasis, amoebiasis and giardiasis), dacarbazine (Hodgkin's disease), tinidazole (metronidazole), ornidazole (antiprotozoal and antibacterial activity), satranidazole (c 10213 go), (trichomoniasis and amoebiasis), cimetidtne (duodenal and gastric ulcers), carbimazole (thyroid disorders), tolazoline (vasodilator action), naphazoline (vasoconstrictor), tetrahydrozoline (vasoconstrictor)[12-14]. Recent studies have been revealed that the substituted

imidazole derivatives attracted attention due to their broad spectrum of pharmacological activities such as anti-inflammatory, analgesic, antimicrobial, antiviral, antifungal, antibacterial, anti-tubercular, anti-cancer, anti-hypertensive, anti-obesity and anti-convulsant[15,16]. In the present studies it has been thought to synthesize newer imidazole derivatives and the structures of the synthesized compounds confirmed on the basis of their elemental analysis and modern analytical techniques such as IR, 1H-NMR and Mass spectral data results.

METHODS

General materials and instrumentations

All the chemicals and solvent procured from E. Merck and S. D. Fine chemicals (India). Melting points were determined by open tube capillary method and are uncorrected. Thin layer chromatography (TLC) plates prepared by silica gel G were used to monitor the reaction as well as to confirm the purity of the compound by using solvent systems as toluene: ethyl acetate: formic acid (5:4:1), benzene: acetone (9:1) were used to run the TLC. The spots were visualized under iodine vapours/UV light. IR spectra were obtained on a Perkin-Elmer 1720 FT-IR spectrometer using KBr pellets. ^{1}H-NMR spectra were recorded on DPX-300 and BRUKER-400 Ultra Shield^{TM} NMR spectrometer, using TMS as internal standard in $CDCl_3$/DMSO-d_6. Microanalysis of the compounds was done on Perkin-Elmer model 240 analyzer and the values were found within ±0.4% of the theoretical values. Mass spectrometry was recorded on LC-MS/MS (WATERS, mass lyns version 4.1) spectrometer.



Compds	R_1	Compds	R_1
3i	H	3vii	4-Br
3ii	4-OH	3viii	4-N(CH_3)$_2$
3iii	2-Cl	3ix	3-OCH_3
3iv	3-Cl	3x	4-OCH_3
3v	3-OH	3xi	2-OH
3vi	4-Cl	3xii	4-F

Figure 1: Synthetic protocol.

Synthesis

The compounds of the synthetic protocol scheme were obtained in the following steps

Synthesis of 1-(Biphenyl-4-yl) ethanone: The starting material biphenyl ethanone (1) was prepared by heating biphenyl with anhydrous $AlCl_3$ in presence of CS_2 and acetic anhydride. The usual work up of the reaction mixture followed by recrystallized from ethanol gave pure compound. The purity of the compound was verified with the help of TLC (B: A, 9: 1). Percentage yield was found 85% and noted Mp.158-160^{O}C. IR spectra are very informative and provided evidence for the formation of the expected structures.

Synthesis of 2-(Biphenyl-4-yl)-2-oxoacetaldehyde: Compound 2-(Biphenyl-4-yl)-2-oxoacetaldehyde (2) was synthesized from biphenyl ethanone (1) in presence of selenium dioxide, usual work up of the reaction mixture gave a yellow liquid which was found pure on TLC examination (TEF 5: 4: 1). The structure of compound was confirmed on the basis of spectral studies.

General procedure for synthesis of 4-(Biphenyl-4-yl)-2-(substituted phenyl)-1*H*-imidazole (3i-xii): Biphenyl-2-oxoacetaldehyde (2) was refluxed with different aromatic aldehyde in presence of ammonium acetate and glacial acetic acid. The usual work up of the reaction mixture followed by recrystallized from acetone to get the desired products (3i-xii). The structures of compounds were confirmed on the basis of their IR and ^{1}H-NMR spectral studies. The compound was found pure on TLC examination (BA 9: 1) and (TEF 5: 4: 1) and its spectral data was found satisfactory for the proposed structures.

Synthesis of 4-(biphenyl-4-yl)-2-phenyl-1H-imidazole (3i): Yield: 70%, Mp: 134-137ºC, R_f =0.53. IR (KBr, cm^{-1}): 3450(C-H, N-H), 3041(C-H, Ar-H), 2871(C-H, CH_2), 1595(C=N), 1564 (C=C). ^{1}H-NMR (DMSO-d_6, δ, ppm): 10.93 (H, s, N-H), 6.53-8.79 (H, m, Ar-H), 7.92 (1H, s, CH, imidazole). ESI-MS (*m/z*): 296 (M^+). Anal.calcd. for $C_{21}H_{16}N_2$: C, 65.11; H, 5.44; N, 9.45. Found: C, 75.11; H, 4.44; N, 9.51.

Synthesis of 4-(4-(biphenyl-4-yl)-1H-imidazol-2-yl) phenol (3ii): Yield: 75%, Mp: 151-154°C, R_f=0.54. IR (KBr, cm^{-1}): 3397(C-H, N-H), 3213(OH), 3035(C-H, Ar-H), 2931(C-H, CH_2), 1675(C=N), 1556 (C=C). ^{1}H-NMR (DMSO-d_6, δ, ppm): 10.51 (H, s, N-H), 8.01-6.79 (H, m, Ar-H), 9.01 (1H, s, CH, imidazole), 9.46 (H, s, OH). ESI-MS (*m/z*): 312 (M^+). Anal.calcd. for $C_{21}H_{16}N_2O$: C, 80.75; H, 5.16; N, 8.97. Found: C, 80.79; H, 4.11; N, 8.95.

Synthesis of 4-(biphenyl-4-yl)-2-(3-chlorophenyl)-1H-imidazole (3iv): Yield: 77%, Mp: 148-151°C, R_f = 0.47. IR (KBr, cm^{-1}): 3405(C-H, N-H), 3021(C-H, Ar-H), 2947(C-H, CH_2), 1623(C=N), 1564 (C=C), 733(C-Cl). ^{1}H-NMR (DMSO-d_6, δ, ppm): 9.97 (H, s, N-H), 7.85-6.59 (H, m, Ar-H), 8.93 (1H, s, CH, imidazole). ESI-MS (*m/z*): 330 (M^+). Anal.calcd. for $C_{21}H_{15}ClN_2$: C, 76.24; H, 4.57; N, 8.47. Found: C, 76.27; H, 4.54; N, 8.37.

Synthesis of 3-(4-(biphenyl-4-yl)-1H-imidazol-2-yl) phenol (3v): Yield: 64%, Mp: 132-133°C, R_f = 0.53. IR (KBr, cm^{-1}): 3451(C-H, N-H), 3267(OH), 3058(C-H, Ar-H), 2879(C-H, CH_2), 1578(C=N), 1551 (C=C). ^{1}H-NMR (DMSO-d_6, δ, ppm): 10.23 (H, s, N-H), 7.95-6.73 (H, m, Ar-H), 8.31 (1H, s, CH, imidazole), 9.37 (H, s, OH). ESI-MS (*m/z*): 312 (M^+). Anal.calcd. for $C_{21}H_{16}N_2O$: C, 64.75; H, 5.16; N, 8.97. Found: C, 60.79; H, 5.21; N, 8.95.

Synthesis of 4-(biphenyl-4-yl)-2-(4-chlorophenyl)-1H-imidazole (3vi): Yield: 72%, Mp: 185-187°C, R_f =0.47. IR (KBr, cm^{-1}): 3379(C-H, N-H), 3086(C-H, Ar-H), 2951(C-H, CH_2), 1663 (C=N), 1491(C=C), 719(C-Cl). ^{1}H-NMR (DMSO-d_6, δ, ppm): 9.73 (H, s, N-H), 7.89-6.57 (H, m, Ar-H), 9.11(1H, s, CH,

imidazole). ESI-MS (*m/z*): 330 (M^+). Anal.calcd. for $C_{21}H_{15}ClN_2$: C, 55.30; H, 4.61; N, 15.71. Found: C, 55.41; H, 4.75; N, 15.92.

Synthesis of 4-(biphenyl-4-yl)-2-(4-bromophenyl)-1H-imidazole (3vii): Yield: 81%, Mp: 142-145°C, R_f = 0.57. IR (KBr, cm^{-1}): 3447(C-H, N-H), 3171 (C-H, Ar-H), 2817 (C-H, CH_2), 1652 (C=N), 1539 (C=C), 803 (C-Br). ^{1}H-NMR (DMSO-d_6, δ, ppm): 10.01 (H, s, N-H), 7.91-6.85 (H, m, Ar-H), 8.71 (1H, s, CH, imidazole). ESI-MS (*m/z*): 375 (M^+). Anal.calcd. for $C_{21}H_{15}BrN_2$: C, 57.21; H, 4.03; N, 7.47. Found: C, 56.79; H, 4.07; N, 7.51.

Synthesis of 4-(biphenyl-4-yl)-2-(4-methoxyphenyl)-1H-imidazole (3x): Yield: 71%, Mp: 160-163°C, R_f = 0.63. IR (KBr, cm^{-1}): 3031(C-H, Ar-H), 2905(C-H, CH_2), 1662(C=N), 1571 (C=C). ^{1}H-NMR (DMSO-d_6, δ, ppm): 9.97 (H, s, N-H), 7.85-6.21 (H, m, Ar-H), 9.03 (1H, s, CH, imidazole), 3.85 (H, s, OCH_3),. ESI-MS (*m/z*): 326 (M^+). Anal. calcd. for $C_{22}H_{18}N_2O$: C, 68.91; H, 5.56; N, 8.51. Found: C, 68.79; H, 5.51; N, 8.58.

Synthesis of 3-(4-(biphenyl-4-yl)-1H-imidazol-2-yl) phenol (3xi): Yield: 78%, Mp: 140-143°C, R_f = 0.55. IR (KBr, cm^{-1}): 3421(C-H, N-H), 3307(OH), 3041(C-H, Ar-H), 2867(C-H, CH_2), 1571(C=N), 1543 (C=C). ^{1}H-NMR (DMSO-d_6, δ, ppm): 10.11 (H, s, N-H), 7.89-6.68 (H, m, Ar-H), 8.73 (1H, s, CH, imidazole), 9.31 (H, s, OH). ESI-MS (*m/z*): 312 (M^+). Anal.calcd. for $C_{21}H_{16}N_2O$: C, 55.45; H, 5.19; N, 9.13. Found: C, 55.51; H, 5.21; N, 9.21.

Synthesis of 4-(biphenyl-4-yl)-2-(4-fluorophenyl)-1H-imidazole (3xii): Yield: 67%, Mp: 171-175 ºC, R_f = 0.45. IR (KBr, cm^{-1}): 3127(C-H, Ar-H), 2945(CH_2), 1635(C=N), 1589(C=C), 861 (C-F). ^{1}H-NMR (DMSO-d_6, δ, ppm): 11.01 (H, s, N-H), 8.11-7.23 (H, m, Ar-H), 9.25 (1H, s, CH, imidazole). ESI-MS (*m/z*): 314 (M^+). Anal.calcd. for $C_{21}H_{15}FN_2$: C, 52.35; H, 4.81; N, 8.91. Found: C, 53.21; H, 4.93; N, 8.97.

Antimicrobial Evaluation

The inhibition of microbial growth under standardized condition may be utilized for demonstrating the therapeutic efficacy of any subtle change in the antibiotic molecule. Which may not be detected by chemical method will be revealed by a reduction in the anti-microbial activity and hence microbiological assays are very useful for resolving doubts regarding possible loss of potency of antibiotics and their preparations of the antibiotic having a known activity. The *in-vitro* antibacterial and antifungal activities of the synthesized compounds were carried out by microdilution susceptibility test using cup-plate technique. Antibacterial activity of newly synthesized compounds (3i-xii) was screened against bacterial strains viz. *Escherichia coli* (*E. coli*, MTCC 2961), *Staphylococcus aureus* (*S. aureus*, MTCC 3160), *Bacillus subtilis* (*B. subtilis*, MTCC 121), *Klebsiella pneumoniae* (*K. pneumoniae*, MTCC 3040) and *Micrococcus luteus* (*M. luteus*, MTCC 7527). The anti-fungal activity was screened against fungal strains viz. *Candida albicans* (*C. albicans*, MTCC 227), *Aspergillus niger* (*A. niger*, MTCC 277) and *Aspergillus flavus* (*A. flavus*, MTCC 418). The MIC (minimum inhibitory concentration) values were determined in compare to standard drug Ciprofloxacin (anti-bacterial) and Fluconazole (anti-fungal). The MIC is considered to be the lowest drug concentration for which there is no microbial growth.

Antibacterial Activity

Experimental Procedure

In-vitro antibacterial activity of the synthesized compounds was tested by disc diffusion method under standard condition using Muller Hinton Agar medium. The test organisms were first cultured in Nutrient broth and incubated for 24 hrs at 37ºC and then freshly prepared bacterial cells were spread onto the Muller Hinton agar plates in a laminar flow cabinet. The test compounds which were previously dissolved in DMSO were then soaked onto sterile discs of Whatman filter paper no. 1 (6 mm diameter). After 24 hrs of incubation at 37ºC, the diameter of zone of inhibition was measured for each compound in mm. The activity was compared with standard antibiotic ciprofloxacin (positive control) and a disc impregnated with dimethylsulfoxide (DMSO) was used as a negative control[17,18]. All the tests were performed in triplicate and the average was taken as final reading. Compounds which have shown good zone of inhibition were selected for minimum inhibitory concentration (MIC) determination.

Antifungal activity

Experimental Procedure

In-vitro antifungal activity of the synthesized compounds was tested by disc diffusion method under standard conditions using Potato dextrose agar medium. Sterile discs of Whatman filter paper no.1 (6 mm diameter) containing specific amounts of an antifungal agent fluconazole (300 mg for the synthesized compounds) were placed on the surface of an agar plate inoculated with a standardized suspension of the microorganisms tested. The plates were incubated at 28±2ºC for 72 hrs for evaluating antifungal activity. A paper disc impregnated with dimethylsulfoxide was utilized as negative control[19,20].

The nutrient agar medium was prepared and autoclaved at 15 lbs pressure for 20 minutes and this media was poured into petri plates and was allowed to solidify. On the surface of media microbial suspension was spread with the help of sterilized cotton swab. Cups were made by boring into agar surface with a previously sterilized cork borer and scooping out the punched part of agar. Four cavities or cups were made in the medium and different concentrations of the test compounds and standard drug Fluconazole were poured in these cavities. The plates were kept at room temperature for 1 hr and then incubated at 37±0.5°C for 24 hrs. The diameter of the zone of inhibition formed around the cavities (cups) after 24 hrs incubation was measured and percentage inhibition of the compound were evaluated. A solvent control was also run to know the activity of the blank[21].

Determination of MIC

MIC of the compound was determined by agar streak dilution method. A stock solution of the synthesized compounds (100 µg/ml) in DMSO was prepared and graded quantities of the test compounds were incorporated in specified quantity of molten sterile agar (Muller Hinton agar).

Table 1: Antibacterial activity measure by zone of inhibition of title compounds (3i-xii).

Compound	*E. coli* (MTCC-1687)		*S. aureus* (MTCC-2940)		*B. subtilis* (MTCC- 441)		*M. luteus* (MTCC 7527)		*K. pneumonia* (MTCC 3040)	
	50 µg/mL±SD[b]	100 µg/ mL±SD	50 µg/ mL±SD	100 µg/ mL±SD	50 µg/mL±SD	100 µg/ mL±SD	50 µg/ mL±SD	100 µg/ mL±SD	50 µg/ mL±SD	100 µg/ mL±SD
3i	8.11±2.35	14.01±2.0	nt	nt	8.32±0.5	9.20±1.3	10.31±1.5	9.31±1.1`	11.30±0.51	12.31±1.15
3ii	14.13±1.71	16.78±1.2	15.61±1.5	17.21±2.5	13.21±1.3	14.21±1.3	14.47±1.3	17.17±1.4	15.41±1.31	18.51±1.37
3iii	12.17±3.26	13.58±1.2	nt	nt	13.51±1.3	15.72±2.3	nt	nt	12.31±1.31	17.31±1.13
3iv	20.31±1.51	23.53±1.2	14.18±1.3	16.87±1.1	19.17±1.1	21.97±0.6	15.21±1.2	16.54±1.1	18.13±1.15	20.53±2.13
3v	15.13±1.33	17.37±1.0	13.17±1.1	14.65±1.7	14.64±1.5	15.61±1.12	13.61±1.5	15.67±0.5	14.22±1.36	14.61±1.13
3vi	14.17±1.12	16.31±1.5	14.61±1.4	15.23±2.5	16.6±1.7	15.21±1.2	11.67±1.5	18.31±1.1	nt	nt
3vii	16.23±2.35	15.31±1.5	nt	nt	15.47±1.2	17.68±1.5	17.21±1.0	18.21±1.0	15.01±1.17	17.12±1.11
3viii	16.15±1.13	17.36±2.3	15.61±1.4	16.23±2.5	14.61±1.5	15.31±1.1	15.17±1.3	17.61±0.2	14.37±1.13	16.89±0.55
3ix	14.15±1.11	16.36±1.3	15.17±1.2	16.33±1.1	13.23±1.1	14.83±1.5	15.93±1.1	17.31±1.2	14.31±1.13	15.17±1.25
3x	18.15±1.41	21.61±1.0	15.11±0.5	17.21± 1.2	17.01±1.31	19.61±1.4	15.16±1.5	14.61±2.1	17.81±1.32	20.61±1.16
3xi	nt	nt	13.61±1.0	15.61±1.3	11.51±1.7	13.12±1.5	9.61±1.1	11.67±2.3	nt	nt
3xii	11.68±1.15	15.51±1.6	11.23±2.0	14.61±1.5	12.33±1.3	14.61±1.5	nt	nt	15.11±1.05	14.58±2.11
Cipro.	27.51±1.21	29.11±1.1	29.31±1.4	30.37±1.7	28.45±1.5	29.81±1.6	28.33±1.5	30.17±1.1	29.41±1.41	30.17±1.35

Measure zone of inhibition in millimeter, SD; Standard Deviation, Compd.; Compunds, Cipro; Ciprofloxacin, nt; means not tested compounds.

Table 2: Antifungal activity as zone of inhibition of title compounds (3i-xii).

Compound	*C. albicans* (MTCC-3617)		*A. niger* (MTCC-281)		*A. flavus* (MTCC 418)	
	50 µg/mL±SD	100 µg/mL±SD	50 µg/mL±SD	100 µg/mL±SD	50 µg/mL±SD	100 µg/mL±SD
3i	15.67±1.53	18.31±1.54	15.67±1.52	19.23±1.08	16.54±1.23	19.13±2.01
3ii	17.12±2.41	19.63±1.21	19.71±1.56	20.19±1.61	19.67±1.53	21.81±1.64
3iii	11.61±1.52	19.17±2.26	17.67±1.52	16.33±1.01	nt	nt
3iv	16.21±1.63	18.73±1.08	19.50±1.12	22.67±1.57	21.50±1.32	23.61±1.53
3v	nt	nt	16.00±1.00	14.31±1.54	15.03±1.42	16.32±1.53
3vi	20.61±1.04	24.21±1.14	21.67±1.08	21.31±1.31	23.67±2.08	13.33±2.53
3vii	14.67±1.63	22.04±1.55	13.33±1.53	16.01±1.06	11.33±1.33	18.56±1.21
3viii	17.31±1.34	19.09±4.65	14.61±1.53	18.83±1.23	21.67±1.51	22.39±1.04
3ix	16.67±2.51	15.31±2.53	16.60±1.53	15.31±2.08	nt	nt
3x	17.13±1.04	19.31±1.53	16.01±1.07	19.36±1.53	18.21±1.22	20.31±1.53
3xi	19.63±3.53	22.20±1.21	13.33±2.51	16.43±1.12	11.31±1.54	17.01±1.32
3xii	nt	nt	16.61±1.15	15.81±1.04	15.61±1.47	16.81±1.01
Fluco.	31.23±1.14	31.81±1.72	30.17±1.32	31.23±1.21	28.56±2.51	31.15±2.13

A specified quantity of the medium (40-50ºC) containing the compound was poured into a Petri dish to give a depth of 3-4 mm and allowed to solidify. Suspension of the micro-organism was prepared to contain approximately 105 cfu/ml and applied to plates with serially diluted compounds in DMSO to be tested and incubated at 37ºC. At the end of the incubation period, the MIC values were determined. All determinations were done in triplicates and the average was taken as final reading. The standard antibiotic, ciprofloxacin (100 µg/ml) used as positive control and 100 mL of DMSO used as a negative control. The MIC was considered to be the lowest concentration of the test substance exhibiting no visible growth of bacteria or fungi on the plate[21].

Statistical analysis

Experimental results were expressed as mean±SD. Student's *t*-test and one-way analysis of variance (ANOVA) were applied to check significant differences in antifungal and antibacterial activities. Differences were considered to be statistically significant at $p < 0.05$.

RESULTS AND DISCUSSION

Chemistry

The title compounds (3i-xii) were synthesized as per synthetic scheme outline. In this scheme biphenyl ethanone (1). Starting material) was treated with selenium dioxide to get 2-(biphenyl-4-yl)-2-oxoacetaldehyde (2). Compound 2 was refluxed with different aromatic aldehydes in presence of ammonium acetate and glacial acetic acid, followed by treatment with chlorobenzene in THF to get twelve new imidazole derivatives (3i-xii). The structures of newly prepared compounds were established on the basis of modern analytical techniques (FT-IR, ^{1}H-NMR and mass spectral data) and elemental analysis.

Structural investigations

The starting material 1-(biphenyl-4-yl) ethanone (biphenyl ethanone, 1) was prepared by heating biphenyl with anhydrous $AlCl_3$ in the presence of CS_2 and acetic anhydride. The usual work up of the reaction mixture followed by recrystallized from ethanol gave pure compound. The purity of the compound was verified with the help of TLC (B: A, 9:1). % age of yield was found 83% and melting point noted 151-152^{0}C. IR spectra are very informative and provided evidence for the formation of expected structures. In general, IR spectra of acetophenone showed a strong band at 1673 cm^{-1} for the confirmation of C=O. Whereas ^{1}H-NMR further confirmed the structure due to the presence of a singlet of CH_3 at 2.61 ppm.

Compound 2 [2-(biphenyl-4-yl)-2-oxoacetaldehyde] was synthesized from 1-(biphenyl-4-yl) ethanone (1) in the presence of selenium dioxide and after work out of reaction gave a yellow liquid which was found pure on TLC examination (TEF 5: 4: 1). The structure of compound was confirmed on the basis of spectral studies. In IR spectra showed a band at 2851 cm^{-1} for the confirmation of aldehydic C-H stretching was very clear. The usual work up of the reaction mixture followed by recrystallized from acetone to get the desired products (3i-xii). The compound was found pure on TLC examination (TEF 5: 4: 1) and its spectral data was found satisfactory for the proposed structures. The structure of this compound was confirmed on the basis of their IR and ^{1}H-NMR spectral studies. In IR spectral studies, the compounds showed intense bands in the region 1535-1633 cm^{-1} of C=N stretching due to the ring closure. In addition, the absorption bands at 1351-1367 cm^{-1} are attributed to the C-N stretching vibrations, which also confirm the formation of desired imidazole ring in the compounds. Whereas ^{1}H-NMR spectra further confirm the structure due to disappearance of the peak of aldehydic proton and appearance of a single peak of NH (imidazole ring) at 11.23 ppm due to ring closure.

Evaluation of Anti-microbial Screening

Anti-bacterial activity

The results of anti-bacterial screening of all the newly synthesized compounds are showed in Table 2 and 3. Compound 3iv [4-(biphenyl-4-yl)-2-(3-chlorophenyl)-1H-imidazole] and compound 3x [4-(biphenyl-4-yl)-2-(4-methoxyphenyl)-1H-imidazole] showed notable activity against *E. coli*, *B. subtilis* and *K. pneumoniae*. Some of them showed moderate activity and others rest good activity. Compounds (3ii, 3viii and 3ix) were shown moderate activity against *E. coli*, *S. aureus*, *M. luteus* and *K. pneumonia*, whereas compounds (3iii, 3vii, 3xi and 3xii) showed mild activity against few bacterial strains µg/ml.

Anti-fungal activity

The results of anti-fungal screening of all the newly synthesized compounds are presented in Table 4 and 5. Compound 3iv [4-(biphenyl-4-yl)-2-(3-chlorophenyl)-1*H*-imidazole] and compound 3x [4-(biphenyl-4-yl)-2-(4-methoxyphenyl)-1*H*-imidazole] were shown notable activity against *E. coli*, *B. subtilis* and *K. pneumoniae*. Some of them showed moderate activity and others rest good activity. Compounds (3ii, 3viii and 3ix), showed moderate activity against *E. coli*, *S. aureus*, *M. luteus* and *K. pneumonia*, while compounds (3iii, 3vii, 3xi and 3xii) showed mild activity against few bacterial strains. The compounds of electron releasing imidazole derivatives (3ii, 3iv, 3viii, 3ix, 3x and xi) presented comparatively better anti-fungal activity than the compounds of electron withdrawing imidazole derivatives (3iii, 3vii and 3xii). Regarding the overall anti-fungal activity was found to be the most potent compound having meta substituted chloro group attached to the aromatic ring and methoxy group on para position[21].

CONCLUSIONS

A number of compounds 3i-xii [4-(biphenyl-4-yl)-2-(substituted phenyl)-*1H*-imidazole] have been successfully synthesized. The pharmacological study was performed to evaluate the effects of substituent on the antibacterial and antifungal activities. The biological activity result revealed that all the newly synthetic compounds 3i-xii [4-(biphenyl-4-yl)-2-(substituted phenyl)-*1H*-imidazole] exhibited better antibacterial activity as compared to antifungal activity in compare to reference drug. The results of anti-

bacterial screening further revealed that among all the compounds, the compound (3iv) and (3x) were observed significant anti-bacterial activity against *E. coli*, *B. subtilis* and *K. pneumoniae* while compounds (3ii), (3viii) and (3ix) as well as compounds (3xi) and (3vii) showed moderate anti-bacterial activity in compare to standard drug ciprofloxacin. The results of anti-fungal screening showed that the compound (3ii) and (3viii) showed good anti-fungal activity against *A. niger* and *A. flavus* and compound (3xi) showed notable activity against *C. albicans*. The compound (3vii) and (3ix) were shown moderate activity against *C. albicans* and *A. niger*.

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AUTHOR'S CONTRIBUTION

Verma BK: writing original draft, methodology, investigation. **Kapoor S:** writing, review and editing, methodology, formal analysis. **Kumar U:** writing, review, and editing, methodology. **Pandey S:** writing, review, and editing, data curation. **Arya P:** writing, review and editing, data curation. Final manuscript was read and approved by all authors.

DATA AVAILABILITY

Data will be made available on request.

CONFLICT OF INTEREST

None to declare.

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