https://ijrps.com

ISSN: 0975-7538
Research Article

Anti-HIV and antimicrobial activity *in vitro* of some new Benzimidazole derivatives

Geeta Yadav*[1], Swastika Ganguly[1], Sankaran Murugesan[2], Abhimanyu Dev[1]

[1]Department of Pharmaceutical Science and Technology, Birla Institute of Technology Mesra, Jharkhand 835 215, India

[2]Department of Pharmacy, Birla Institute of Technology & Science, Pilani, Rajasthan 333 031, India

ABSTRACT

A series of 2-(1H-benzo[d]imidazol-1-yl)-N-4-aryl acetamides (**11-20**) were synthesized and evaluated to investigate their possible anti-HIV and antimicrobial activities. HIV-1 RT inhibitory activity was evaluated by using HIV-1 RT RNA-dependent DNA polymerase activity assay. Among these derivatives, compounds **12** and **20** were found to have reasonable HIV-1 RT inhibitory activity while the rest exhibited weak activity in comparison to the standard efavirenz. On the other hand, most of the test compounds were found to be significantly effective against gram positive bacteria (*Staphylococcus aureus* and *Bacillus subtilis*) and some gram-negative bacterial strains (*E.coli, Salmonella typhi*, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa)*, among which compounds **12, 13, 17 and 19** sprouted out as the most effective antibacterials but less effective than the standard ciprofloxacin. In case of fungal strains like *Candida albicans* and *Aspergillus niger*, almost all the compounds were found to exhibit potency comparable or more than that of standard drug fluconazole except for the compound **11** which exhibited moderate to good activity against both the fungal strains. For a deep insight into the role of different functional groups at different positions of phenyl ring attached with the help of a linker group to the N-1 position of benzimidazole, structure activity relationship (SAR) study was carried out, which clearly states the importance of chloro and methyl groups in enhancing the antimicrobial activity. ADME prediction study also indicates the significant drug like properties of the derivatives.

Keywords: Benzimidazole; Antibacterial activity; Antifungal activity; Anti-HIV activity; SAR; ADME

INTRODUCTION

AIDS comes under the category of most gruesome diseases, which according to WHO reports affected near about 33 million people in 2008 (UNAIDS/WHO report, 2008) and engulfed approximately 25 million AIDS-related people in the last 25 years (UNAIDS/WHO report, 2007). Human immunodeficiency virus (HIV) belongs to the lentivirus family, mainly responsible for AIDS/HIV (Gonda *et al,* 1985, Gonda *et al,* 1986). Up to an extent anti-retroviral therapy (HAART) succeeded in its aim to rein the disease but the ever increasing problem of resistance development against drugs of this therapy has become a matter of great worry especially in context with the developing countries, where still HIV is a major health and socioeconomic issue (Ghosh *et al,* 1986).

Another one of the critical problems embedded worldwide nowadays, is the infectious microbial diseases. For the treatment of microbial diseases generally antimicrobials are used. Antimicrobial agents kill the invading microorganisms without harming the host cells. The concentration of antimicrobials is carefully controlled in such a manner that it only attacks the microbial cells and well tolerated by the host cells (Rang *et al,* 2003). However, the resistance development against antimicrobials among the significant species of microorganisms (Gram-positive bacteria and some fungi) is a major serious health concern. A big share of the affected population belongs to the developing countries because of the non-availability of desired drug entities (Sharma *et al,* 2009). Need of the hour is the development of some novel highly resistant barrier drugs and also the careful use of present or existing antimicrobial agents due to cross-resistance problem. (He *et al,* 2003; Becker *et al,* 2006; Metwally *et al,* 2006).

Increasing risk of bacterial and fungal resistance development has forced the researchers and academicians to adopt some new strategies to combat this critical resistance problem. The ultimate challenge is to synthesize some new drugs with novel mechanism, low systemic side effects, high resistance barrier, enhanced biological activity profile and chemical properties different from the existing ones. Though, there are a large

* Corresponding Author
Email: yadavgeeta172@gmail.com
Contact: +91-8986672366
Received on: 20-09-2014
Revised on: 17-11-2014
Accepted on: 19-11-2014

numbers of antimicrobial drugs in stock yet there will always be a vital need to explore new agents and exploit the old ones due to antimicrobial resistance (Fidler *et al,* 1998; Macchiarulo *et al,* 2002).

Benzimidazole nucleus came into existence naturally as a part of vitamin B_{12} (Barker *et al,* 1960) and bears structural similarity with the naturally occurring nucleotides, which helps it to interact more easily with the biophores in the microorganisms (Starcevic *et al,* 2007). Benzimidazoles remain one of the most versatile classes of compounds against microbes and, therefore, need to be explored more. Extensive literature survey also revealed that azole drugs, specifically fused imidazoles exhibit a number of pharmacological activities such as antiviral, anticancer, antiulcer, antimicrobial and anthelmintic activities (Kumar *et al,* 2006; Refaat 2010; Kilcig and Altanlar 2006; Kerimov *et al,* 2007). There are several clinically approved drugs in the market having this fused azole moiety like omeprazole, mebendazole, pimobendan and albendazole. In this communication and in continuation of our *in silico* studies on the N-1 substituted benzimidazoles (Gangu-ly and Yadav, 2013) we wish to report here the synthesis of some benzimidazole derivatives containing alkyl spacer group and substituted acetamido groups. These compounds were evaluated for their HIV-1-RT inhibitory and antimicrobial activities.

MATERIAL AND METHODS

CHEMISTRY

All reagents were purchased from commercial suppliers like Sigma Aldrich, Merck India Ltd., Himedia and Rankem chemicals and were of GR or AR grade. The purity of the compounds were assessed by monitoring TLC performed on Merck silica gel 60 F254 aluminium sheets and TLC spots were seen using Iodine chamber and Shimadzu (UV-254) spectrometer. Melting points were recorded using an Opti-melting point automatic apparatus and were uncorrected. IR spectra (KBr disc / or pallets) were recorded on SHIAMADZU FT / IR 8400 and were reported in cm^{-1}. ^{1}H-NMR Spectra were recorded on BRUKER Advance Digital Spectrophotometer 400 MHz. Chemical shifts are expressed in δ values (ppm) relative to TMS as an internal standard, using DMSO-d6 and The mass spectrawere recorded on a Jeol SX-102 instrument.

EXPERIMENTAL

Methods of synthesis

Synthesis of 2-chloracetamides (1-10)

General procedure

Appropriate primary aromatic amines were treated with 2-chloroacetylchloride resulting in the formation of corresponding chloroacetamides. The reaction was carried out in ice-bath for 30 min and then at room temperature for 1 h (Soyer *et al,* 2004).

Synthesis of 2-(1H-benzo[d]imiazol-1-yl)-N-aryl chloracetamides (11-20)

General method

To a solution of benzimidazole (0.012 M) and appropriate 2-chloro-N-aryl acetamide (0.012 M) in 20 ml of Dimethyl formamide (DMF), K_2CO_3 (0.024 M) was added with stirring. The mixture was refluxed for 14-16 h. After that, the mixture was cooled, poured into crushed ice to yield a precipitate which was filtered, washed with water (3x100 ml) and finally with methanol. Recrystallization was performed with ethanol to yield the title compounds.

2-(1H-benzo[d]imidazol-1-yl)-N-4-tolyl acetamide (11) (11)

Yield: 88%, M.P. 222-224^{0}C, IR (KBr):3284.77 (NH stretching), 2792.2 (C-H stretching), 1668.43 (C=O stretching), 1346.31 (C-N aromatic stretching), 1192.01 (C-N aliphatic stretching)cm^{-1}; ^{1}H NMR (DMSO-d6, 400MHz): 2.19 (s; 3H; CH_3), 4.72 (s; 2H; CH_2) 6.81-7.90 (m; 8H; Ar-H), 9.98 (s;1H; NH); EI-MS:266 (M^++ 1).

2-(1H-benzo[d]imidazol-1-yl)-N-(2,4-dimethylphenyl) acetamide (12)

Yield: 86%, M.P. 235-237^{0}C, IR (KBr): 3360 (NH stretching), 1685.84 (C=O stretching), 1329 and 1303.92 (C-N aromatic stretching), 1238.34 and 1209.41 (C-N aliphatic stretching), 2955.04, 2841.24, 1458.23 and 1396.51 (C-CH_3 stretching)cm^{-1}; ^{1}H NMR (DMSO-d6, 400MHz): 2.46(s; 3H; CH_3) 2.17(s; 3H; CH_3) 2.98 (s; 2H; CH_2) 4.52 (s; 2H; CH_2) 6.86-7.80 (m; 7H; Ar-H), 8.13 (s;1H; =CH), 9.21 (s;1H; NH) ; EI-MS:280 (M^++ 1).

2-(1H-benzo[d]imidazol-1-yl)-N-(3,4-dimethylphenyl) acetamide (13)

Yield: 75%, M.P. 208-210^{0}C, IR (KBr): 3267.06 (NH stretching), 1670.85 (C=O stretching), 1309.98 and 1259.87 (C-N aromatic stretching), 1176.22 and 1126.78 (C-N aliphatic stretching), 3061.96, 2928.87 and 1400 (C-CH_3 aralkyl stretching)cm^{-1}; ^{1}H NMR ((DMSO-d6, 400MHz): 2.35 (s; 3H; CH_3) 2.35 (s; 2H; CH_3) 5.14 (s; 2H; CH_2) 7.05-7.95(m; 7H; Ar-H), 8.15 (s;1H; =CH), 10.38(s;1H; NH); EI-MS:280 (M^++ 1).

2-(1H-benzo[d]imidazol-1-yl)-N-(4-ethylphenyl) acetamide (14)

Yield: 85%, M.P. 185-187^{0}C, IR (KBr): 3244.38 (NH stretching), 1662.69 (C=O stretching), 1263.42 (C-N aromatic stretching), 1205.55 and 1031.95 (C-N aliphatic stretching), 2937.68 and 1450.52 (C-CH_3 stretching)cm^{-1}; ^{1}H NMR (DMSO-d6, 400MHz): 1.25(s; 3H; CH_3) 2.58(s; 2H; CH_2) 5.12 (s; 2H; CH_2) 7.05-7.80 (m; 8H; Ar-H), 8.15 (s;1H; =CH), 10.45(s;1H; NH); EI-MS:280 (M^++ 1).

2-(1H-benzo[d]imidazol-1-yl)-N-(3-methoxyphenyl) acetamide (15)

Yield: 76%, M.P. 205-207^{0}C, IR (KBr): 3377.47 (NH stretching), 1687.77 (C=O stretching), 1307.78 (C-N aromatic stretching), 1220.98 (C-N aliphatic stretching), 1251.84, 1020.98 and 1237.38 (C-O aralkyl stretching) cm^{-1}; ^{1}H NMR (DMSO-d6, 400MHz): 3.68(s; 3H; CH_3) 4.66 (s; 2H; CH_2) 6.18-7.79 (m; 9H; Ar-H), 9.85 (s;1H; NH); EI-MS:282 (M^++ 1).

2-(1H-benzo[d]imidazol-1-yl)-N-(4-methoxyphenyl) acetamide (16)

Yield: 79%, M.P. 204-205^{0}C, IR (KBr): 3255.84 (NH stretching), 1662.64 (C=O stretching), 1307.74 and 1249.87 (C-N aromatic stretching), 1249.87 and 1186.22 (C-N aliphatic stretching), 1249.87, 1033.85 and 1186.32 (C-O aralkyl stretching)cm^{-1}; ^{1}H NMR (DMSO-d6, 400MHz): 3.71(s; 3H; CH_3) 5.13 (s; 2H; CH_2) 6.88-8.22 (m; 9H; Ar-H), 10.32 (s;1H; NH); EI-MS:282 (M^++ 1).

2-(1H-benzo[d]imidazol-1-yl)-N-(2-chlorophenyl) acetamide (17)

Yield: 86%, M.P. 180-183 ^{0}C, IR (KBr): 3134.43 (NH stretching), 1716.70 (C=O stretching), 1350 and 1250 (C-N aromatic stretching), 1150 and 1049.31 (C-N aliphatic stretching), 3700 (OH phenolic stretching), 1200 and 790.30 (C-Cl stretching).cm^{-1}; ^{1}H NMR (DMSO-d6, 400MHz): 4.88 (s; 2H; CH_2) 6.94-8.03 (m; 9H; Ar-H), 10.24 (s;1H; NH) ; EI-MS:286 (M^++ 1).

2-(1H-benzo[d]imidazol-1-yl)-N-(4-ethoxyphenyl) acetamide (18)

Yield: 79%, M.P. 215-218^{0}C, IR (KBr): 3331.18 (NH stretching), 1681.98 (C=O stretching), 1249.91 and 1292.35 (C-N aromatic stretching), 1213.27 and 1193.98 (C-N aliphatic stretching), 1249.91, 1213.27, 1043.52 and 1168.90 (C-O aralkyl stretching) cm^{-1}; ^{1}H NMR ((DMSO-d6, 400MHz): 1.30(s; 3H; CH_3) 3.98 (s; 2H; CH_2) 5.10(s; 2H; CH_2) 6.85-7.98 (m; 8H; Ar-H), 8.16 (s;1H; =CH), 10.28(s;1H; NH); EI-MS:296 (M^++ 1).

2-(1H-benzo[d]imidazol-1-yl)-N-(2-chloro-6-methylphenyl) acetamide (19)

Yield: 78%, M.P. 247-248^{0}C, IR (KBr): 3302.24 (NH stretching), 1683.91 (C=O stretching), 1350 (C-N aromatic stretching), 1190.12 and 1066.67 (C-N aliphatic stretching), 739.30 (C-Cl stretching)cm^{-1}; ^{1}H NMR (DMSO-d6, 400MHz): 2.14(s; 3H; CH_3) 5.48 (s; 2H; CH_2) 7.01-7.83 (m; 8H; Ar-H), 9.48 (s;1H; NH) ; EI-MS:300 (M^++ 1).

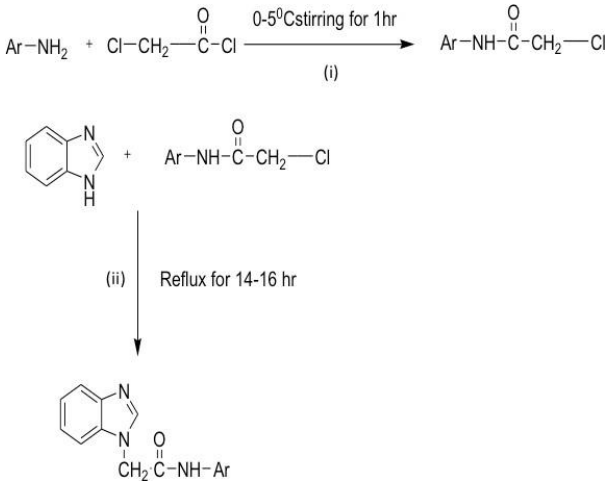
2-(1H-benzo[d]imidazol-1-yl)-N-(4-chloro-2-methylphenyl) acetamide (20)

Yield: 68%, M.P. 245-246^{0}C, IR (KBr): 3202.26 (NH stretching), 1697.34 (C=O stretching), 1320 (C-N aromatic stretching), 1276.70 (C-N aliphatic stretching), 1481.34 and 757.30 (C-Cl stretching)cm^{-1}; ^{1}H NMR(DMSO-d6, 400MHz): 2.16(s; 3H; CH_3) 4.73 (s; 2H; CH_2) 6.67-7.81 (m; 8H; Ar-H), 9.20 (s;1H; NH) ; EI-MS:300 (M^++ 1).

RESULTS AND DISCUSSION

Chemistry

According to reported procedure chloracetamides were synthesized by the reaction of different appropriate primary aromatic amines with 2-chloroacetylchloride resulting in the formation of corresponding 2-chloroacetamides (Soyer *et al,* 2004). Confirmation of the compounds **1-10** was done by physical and spectral data of the reported compounds while the structures of the final compounds (**11-20**) were confirmed by their spectral data (IR, ^{1}H NMR as well as mass analysis) Compounds 11-20 were prepared by the method reported for benzimidazoles (Rajput and Gore, 2012: Rajput and gore, 2010; Popkov et al, 2008). Benzimidazole on treatment with the appropriate 2-chloroacetamides in the presence of potassium carbonate yielded the N-1 substituted benzimidazoles **11-20**. The reaction sequence for different synthesized compounds is layed out in Scheme 1.



Scheme 1: Synthesis of benzimidazole derivatives

Reagents and conditions: (i) glacial acetic acid, 0-5°C, 1h (ii) DMF, reflux 14-16 h

11:Ar=4-$CH_3C_6H_4$	16:Ar=4-$OCH_3C_6H_4$
12:Ar=2,4-$CH_3C_6H_3$	17:Ar=2-ClC_6H_4
13:Ar=3,4-$CH_3C_6H_3$	18:Ar=4-$OC_2H_5C_6H_4$
14:Ar=4-$C_2H_5C_6H_4$	19:Ar=2-Cl,6-$CH_3C_6H_3$
15:Ar=3-$OCH_3C_6H_4$	20:Ar=4-Cl,2-$CH_3C_6H_3$

In-vitro HIV-1 RT inhibitory activity

Synthesized compounds **11-20** were evaluated for HIV-1 RT inhibitory activity by using HIV-1 RT RNA-dependent DNA polymerase activity assay (Cremer *et al,* 2006). Efavirenz was taken as standard drug for comparison. Results obtained for the activity are shown in Table 1.

Antimicrobial activity test

All the synthesized compounds were also evaluated for their *in vitro* antibacterial activity against Gram-positive bacteria: *Staphylococcus aureus* (NCIM 2122), *Bacillus subtilis* (MTCC 121), Gram-negative bacteria:

Table 1: HIV-1 RT inhibitory activity of synthesized compounds

S.No	Compound code	% RT inhibition (20µM)
1	11	22.4
2	12	51.8
3	13	22.19
4	14	23.9
5	15	11.98
6	16	22.31
7	17	37.82
8	18	11.9
9	19	23.9
10	20	49.54
11	Efavirenz	97.76

Table 2: Antibacterial activity of compounds 11-20 (MIC in µg/mL)

Compounds	Microorganisms					
	Gram +ve bacteria		*Gram -ve bacteria*			
	S. aureus	*B.subtilis*	*E. coli*	*P.aeruginosa*	*S. typhi*	*K. pneumoniae*
11	12.5	12.5	25	12.5	25	25
12	3.125	6.25	3.125	6.25	6.25	3.125
13	3.125	6.25	6.25	6.25	6.25	6.25
14	6.25	12.5	12.5	6.25	12.5	6.25
15	12.5	12.5	25	25	25	25
16	6.25	6.25	12.5	12.5	25	12.5
17	3.125	3.125	6.25	6.25	12.5	6.25
18	6.25	6.25	12.5	6.25	6.25	12.5
19	3.125	3.125	6.25	6.25	6.25	6.25
20	6.25	3.125	12.5	6.25	6.25	12.5
Ciprofloxacin	≤1	≤1	≤1	≤1	≤1	≤1

Escherichia coli (MTCC118), *Pseudomonas aeruginosa* (MTCC 647), *Salmonella typhi* (NCIM 2501), *Klebsiella pneumonia (*MTCC 3384) and fungus *Candida albicans* (MTCC 227), *Aspergillus niger* (NCIM 1056), by using the two fold serial dilution technique and the results obtained are shown in Table 3. For antibacterial and antifungal activity ciprofloxacin and fluconazole were used as the standard drugs respectively.

Table 2 and 3: In *vitro* antibacterial and antifungal activity data of test compounds respectively

ADME prediction

QikProp tool is generally used by the researchers for ADME analysis of lead compounds (Schrodinger Inc. 2008). This software not only predicts the physico-chemical parameters but also its pharmacokinetic properties and compounds also have to go through Lipinski's rule of five (Lipinski *et al,* 2001; Lipinski *et al,* 1997). So, ADME properties of benzimidazole drivatives (**11**-**20**) were predicted by using maestro Qikprop v3.0 tool and results are concluded in Table 4.

HIV-1 RNA-dependent DNA polymerase activity assay

"Poly (rA)/oligo(dT) was used as a template for the RNA-dependent DNA polymerase reaction by HIV-1 RT, either wild type or carrying the mutations. For the activity assay, 25 µl final reaction volume contained TDB buffer (50 mM Tris-HCl (pH 8.0), 1 mM dithiothreitol, 0.2 mg/ml bovine serum albumin, 2% glycerol), 10 mM MgCl2, 0.5 mg of poly(rA)/oligo(dT)10:1 (0.3 mM 3′-OH ends), and 10 mM 3[H]-dTTP (1 Ci/mmol), and was introduced into tubes containing aliquots of different enzyme concentrations (5 to 10 nM RT). After incubation at 37°C for indicated time, 20 µL from each reaction tube were spiked on glass fiber filters GF/C and immediately immersed in 5% ice-cold trichloroacetic acid (TCA) (AppliChem GmbH, Darmstadt). Filters were washed three times with 5% TCA and once with ethanol for 5 min, then dried, and finally added with EcoLume scintillation cocktail (ICN, Research Products Division, Costa Mesa, CA, USA) to detect the acid precipitable radioactivity by PerkinElmer Trilux MicroBeta 1450 Counter (Waltham, MA, USA)" (Cremer *et al,* 2006).

Microbiology

Minimum inhibitory concentration (MIC) of the synthesized compounds was carried out by using two fold serial dilution method (Collins *et al,* 2004; Kus *et al,* 2009). The targeted compounds were dissolved in dimethyl sulfoxide (DMSO) to make a stock solution of 1000µg/ml concentration. From this stock solution dilutions were prepared with the Mueller-Hinton agar medium of 100µg/ml, 50 µg/ml, 25 µg/ml, 12.5 µg/ml, 6.25 µg/ml and 3.125 µg/ml. Test tubes containing the

Table 3: Antifungal activity of compounds 11-20 (MIC in µg/mL)

Compounds	Microorganisms	
	C.albicans	*A.niger*
11	6.25	12.5
12	3.125	3.125
13	3.125	3.125
14	6.25	6.25
15	6.25	3.125
16	3.125	3.125
17	3.125	3.125
18	3.125	6.25
19	3.125	3.125
20	6.25	6.25
Fluconazole	12.5	12.5

Table 4: Prediction of ADME properties of designed benzimidazole analogs using Qikprop v3.0 tool

Compd. no.	Mol MW	Log Po/w	Log S	Log BB	PMD CK	Human oral absorption (%)	Rule of five
11	265.31	3.332	-4.063	-0.269	1333.861	100	0
12	279.34	3.638	-4.627	-0.262	1334.064	100	0
13	279.34	3.696	-4.751	-0.325	1219.945	100	0
14	279.34	3.618	-4.800	-0.261	1211.619	100	0
15	281.31	3.133	-4.012	-0.219	1220.145	100	0
16	281.31	3.129	-4.007	-0.268	1048.792	100	0
17	285.73	3.531	-4.553	-0.393	2591.667	100	0
18	295.34	3.576	-4.665	-0.273	1220.754	100	0
19	299.75	3.598	-4.268	-0.373	2797.867	100	0
20	299.75	3.780	-4.983	-0.376	2622.397	100	0

only the culture medium and DMSO in the same dilu---tions were used as control and 10^4 CFU/ml concentra---tion of microbial strains was used during the screening. The MIC values were recorded incubating the test tubes at 35^0C for 24 hrs. For antibacterial activity ciprofloxacin was taken as a reference drug.

Antifungal activity of the synthesized compounds was also calculated by using the same method as used for bacteria. But, In case of fungus sabouroud dextrose broth was taken as media and MIC values were also taken after incubation for 48 h at 28±2°C. For antifun---gal activity fluconazole was used as a standard drug. Each experiment was replicated twice to calculate the MIC values.

ADME properties

ADME studies help in the prediction of pharmacokinet---ic characteristics, metabolic pathways and in the de---sign of *in vivo* studies of a drug. The significant de---scriptors like octanol/water partition coefficient Clog P, log S, log BB, PMDCK, human oral absorption (%) and rule of five of the compounds have been calculated using maestro Qikprop v3.0 tool of maestro, for pre---dicting the drug-like properties of molecules. These properties are Molecular weight (mol MW), Oc---tanol/water partition coefficient (Log Po/w), Aqueous solubility (QPlogS), Brain/blood partition coefficient (QPlogBB), MDCK cell permeability (QPPMDCK) and percent human oral absorption.

QP log Po/w predicts the partition coefficient, which is important for the estimation of absorption and distri-bution of drugs within the body. For the lead com-pounds, the partition coefficient (QPlogP o/w) and wa---ter solubility QPlogS), critical for estimation of absorp---tion and distribution of drugs within the body ranged between 3.129 to 3.780 and −4.007 to −4.983.Blood brain barrier permeability (QPlogBB), a key factor gov---erning drug access to brain, ranged from -0.219 to -0.376 and QPPMDCK value ranges from 1048.792 to 2797.867. Overall, the percentage human oral absorp---tion for the compounds reported was predicted to be 100 %.

STRUCTURE ACTIVITY RELATIONSHIP (SAR) STUDY

Among the synthesized analogs, **12 and 20** surfaced out with moderate *in vitro* HIV-1 RT inhibition in the range of 50-52 % while remaining of the compounds resulted in weak HIV-1 RT inhibitory activity at 20 µM concentrations. Compound **12** and **20** substituted with Cl and CH_3 group at different positions of the phenyl ring attached to the spacer group at the N-1 position of the benzimidazole moiety showed approximately 50% inhibition of HIV-1 RT. It was interesting to pen down here that lipophilic character of the substituents, par---ticularly halogens and methyl groups may be account---able for the reasonable HIV-1- RT inhibitory activity.

In case of anti-bacterial activity, the compounds **12**, **13**, **17** and **19** portrayed highest activity almost against all

the bacterial strains (Gram-positive and Gram-negative) while compound **20** also displayed somewhat significant activity. The other compounds showed moderate activity towards all the bacterial strains. However, overall the activity was less than that of standard drug ciprofloxacin. It was noteworthy to report here that synthesized analogs showed more sensitivity towards Gram-positive bacteria than the Gram-negative bacteria.

From the results of anti-microbial activity of the newly synthesized compounds (**11-20**), it was apparent that lipophillicity is an important factor which affects the antibacterial activity as is evident for the compounds **12**, **13**, **16** and **17**. This was also authenticated by the fact that a decrease in the lipophillicity led to a significant drop in activity as evident for compound **11**. A combination of halogen and methyl substitution in the various positions of the aromatic ring also showed very good antibacterial activity which is evident from compound **19**. Among the screened compounds, compound **17** bearing electron withdrawing group such as chloro at 2-position of the phenyl ring attached at the N-1 position of benzimidazole through a spacer group, exhibited prominent antibacterial activity against both gram positive bacteria *S. aureus and B. subtilis,* while showed moderate activity against *K. pneumonia, E.Coli and P.aeruginosa* and mild activity towards *S.typhi*.

In case of antifungal activity, almost all the compounds were found to exhibit potency comparable or more than that of standard drug fluconazole against both the fungal strains *C.albicans* and *A.niger*. In fact, the activity was higher than that of standard fluconazole while the compound **11** showed equipotent activity as fluconazole against *A.niger* fungal strain while higher in case of *C.albicans*. Compounds **12**, **13**, **16**, **17** and **19** were found to have more noteworthy antifungal activity in comparison to the standard drug and rest of the compounds of the series. Primarily electron donating groups showed excellent fungicidal activity against the fungal strains *C. albicans* and *A. niger.*

For the prediction of the dug likeness of the novel benzimidazoles, some physicochemical parameters and the pharmacokinetically relevant properties of the target compounds were calculated using maestro Qikprop v3.0 tool. For this purpose, different properties like log Po/w, log S, log BB, PMDCK, human oral absorption (%) and rule of five of the compounds were determined as indicated in (Table 4) and the results were found to have drug like properties of satisfactory level.

CONCLUSION

Some N-1-substituted 2-(1H-benzo[d]imiazol-1-yl)-N-aryl acetamides **11-20** were synthesized and evaluated for *in vitro* anti-HIV and antimicrobial activities. In addition, to predict drug like properties, ADME studies were also carried out. For a deep insight, Structure activity relationship studies were performed which revealed the fact that a combination of an optimal hydrophobicity with electron donating or electron withdrawing groups at various positions of the phenyl ring attached to the benzimidazole nucleus at the N-1 position may be important for achieving significant antibacterial and antifungal activities. In case of HIV-1-RT inhibitory activity, a prevalence of electronegative groups such as halogens was responsible for the moderate activity. An ADME study of all the synthesized compounds has shown that all the compounds possess drug like properties. Findings from the SAR and ADME studies indicate that these newly synthesized benzimidazoles may further act as lead compounds.

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

ACKNOWLEDGMENTS

The authors acknowledge University Grants Commission for providing financial support in the form of a Major Research Project. One of the authors (GY) gratefully acknowledges the University Grants Commission-Basic Science Research (UGC-BSR) for the award of fellowship during the work.

REFERENCES

Barker, H.A., Smyth, R.D., Weissbach, H., Toohey, J.I., Ladd, J.N., Volcani, B.E. Isolation and properties of crystalline cobamide coenzymes containing benzimidazole or 5, 6-dimethylbenzimidazole. Journal of Biological Chemistry. vol.235, no.2, 1960 pp. 480-488.

Becker, K., Hu, Y., Biller-Andorno, N. Infectious diseases–a global challenge. International Journal of Medical Microbiology. vol. 296, no.4, 2006 pp. 179–185.

Collins, C.H., Lyne, P.M., Grange, J.M., Falkinham, J.O. Collins & Lyne's Microbiological Methods. 8th ed., Hodder Arnold Publication: Butter-worth, London. 2004

Cremer, N.S., Hamamouch, N., Felix, A.S., Velazquez, S., Balzarini, J., Camarasa, M.J. Structure activity relationship of [2″,5″-bis-O-(tert-butyl dimethyl silyl)-β-D-ribofuranosyl]-3″-spiro-5-(4-amino-1,2-oxathiole-2,2-dioxide) thymine derivatives as inhibitors of HIV-1 reverse transcriptase dimerization. Journal of Medicinal Chemistry. vol.49, 2006 pp. 4834–4841.

Fidler, D.F. Legal issues associated with antimicrobial drug resistance. Emerging Infectious Diseases. vol. 4, no.2 1998 pp. 169–177.

Ganguly, S.; Yadav, G. Molecular Docking Studies of Novel Benzimidazole Analogs as HIV-1-RT Inhibitors with Broad Spectrum Chemotherapeutic Properties, Int. J. Drug Design and Dis., **2013**, *4*, 1193-1213.

Ghosh, T.K. AIDS: a serious challenge to public health. Journal of the Indian Medical Association.vol.84, no.1, 1986 pp. 29-30.

Gonda, M.A., Braun, M.J., Clements, J.E., Pyper, J.M, Wong-Staal, F., Gallo, R.C and Gilden, R.V. Human T-cell lymphotropic virus III shares sequence homology with a family of pathogenic lentivirus. ; Proceedings of the National Academy of Sciences of the United States of America. vol. 83, 1986 pp. 4007-4011.

Gonda, M.A., Wong-staal, F, Gallo, R.C., Clements, J.E., Narayan, O., and Gilden, R.V. Sequence homology and morphologic similarity of HTLV-III and visna virus, a pathogenic lentivirus. Proceedings of the National Academy of Sciences of the United States of America.vol. 227, 1985 pp. 173-177.

He, Y., Wu, B., Yang, J., Robinson, D., Risen, L., Ranken, R., Blyn, L., Sheng, S., Swayze, E.E. 2-Piperidin-4-yl-benzimidazoles with broad spectrum antibacterial activities. Bioorganic Medicinal Chemistry Letters. vol.13, no.19, 2003 pp. 3253–3256.

Kerimov, I., Kilcgil, A.G., Eke, B.C., Altanlar, N., Iscan, M.J. Synthesis, antifungal and antioxidant screening of some novel benzimidazole derivatives. Journal of Enzyme Inhibition of Medicinal Chemistry. vol.22, no.6, 2007 pp. 696-701.

Kilcig, G.A., Altanlar, N. Synthesis and antifungal activity of some benzimidazole derivatives.Turkish Journal of Chemistry. vol. 30, 2006 pp. 223-228.

Kumar, B.V.S., Vaidya, S.D., Kumar, R.V., Bhirud, S.B., Mane, R.B. Synthesis and anti-bacterial activity of some novel 2-(6-fluorochroman-2-yl)-1-alkyl/acyl/aroyl-1H-benzimidazoles. European Journal of Medicinal Chemistry. vol. 41, no.5, 2006 pp. 599-604.

Kus, C., Sozudonmez, F., Altanlar, N. Synthesis and antimicrobial activity of some novel 2-[4-(Substituted piperazin/piperidin-1-ylcarbonyl) phenyl]-1Hbenzimidazole derivatives. Archiv der Pharmazie. vol. 342, no.1, 2009 pp.54-60.

Lipinski, C.A., Lombardo, F., Dominy, B.W., Feeney, P.J. Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. Advanced Drug Delivery Review. vol. 23, 1997 pp. 3–25.

Lipinski, C.A., Lombardo, F., Dominy, B.W., Feeney, P.J. Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. Advanced Drug Delivery Reviews. vol.46, 2001 pp. 3–26.

Macchiarulo, A., Constantino, G., Fringuelli, D., Vecchiarelli, A., Fausto, S., Renata, F. 1,4-Benzothiazine and 1,4-Benzoxazine imidazole derivatives with antifungal activity: A docking study. Bioorganic Medicinal Chemistry. Vol.10, no.11, 2002 pp. 3415–3423.

Metwally, K.A., Abdel-Aziz, L.M., Lashine, E.M., Husseiny, M.I., Badawy, R.H. Hydrazones of 2-aryl-quinoline-4-carboxylic acid hydrazides: synthesis and preliminary evaluation as antimicrobial agents. Bioorganic Medicinal Chemistry. vol. 14, no. 26, 2006 pp. 8675-8682.

Popkov, S.V; Alekseenko, A.L; Lobanova, E.G; Zhurav---lev, V.N. Preparation of substituted w-azolylalkananilides as platelet aggregation inhibitors and process for their preparation. Russ. **2008**, RU2322440C120080420.

Rajput, A.P.;Gore, R.P. N-Alkylation of azoles disubstituted acetamides under microwave and mannich re---action conditions, Pharma Chemica, **2012**, *6*, 2222-2227.

Rajput, A.P.;Gore, R.P. Synthesis and characterization of some new azole-acetanilides, J.Chem. Pharm. Res., **2010**, *5*, 52-59.

Rang, H.P., Dale, M.M., Ritter, J.M and Flower, R.J. Analgesic drugs, Rang and Dale's Pharmacology (5th edn). Churchill Livingstone. 562, 2003.

Refaat, H.M. Synthesis and anticancer activity of some novel 2-substituted benzimidazole derivatives. European Journal of Medicinal Chemistry. vol. 45, no.7, 2010 pp. 2949-56.

Schrodinger Inc. 2008, USA, Schrodinger, LLC, New York (accessed on 15 August 2014).

Sharma, D., Narasimhan, B., Kumar, P., Judge, V., Narang, R., De Clercq, E., Balzarini, J. Synthesis, antimicrobial and antiviral evaluation of substituted imidazole derivatives. European Journal of Medicinal Chemistry. vol.44, no.6, 2009 pp. 2347–2353.

Soyer, Z., Kilic. F.S., Erol, K., Pabuccuoglu, V. Synthesis and anticonvulsant activity of some ώ-(1H-imidazol-1-yl) -N-phenylacetamide and propionamide derivatives. IL Farmaco vol.59, no.8, 2004 pp. 595-600.

Starcevic, K., Kralj, M., Ester, K., Sabol, I., Grace, M., Pavelic, K., Zambola, G.K. Synthesis, antiviral and antitumor activity of 2-substituted-5-amidino-benzimidazoles. Bioorganic Medicinal Chemistry. vol.15, no.13, 2007 pp. 4419–4426.

UNAIDS/WHO (2008). Report on the global AIDS epidemic. Retrieved from http: //www.unaids.org/en/ KnowledgeCenter/HIVData/GlobalReport/2008/2008 Global report.asp. Accessed on august 24, 2014.

UNAIDS/WHO(2007). AIDS epidemic update. Retrieved from http://www.data.unaids.org. Accessed on august 24, 2014.