Academic Sciences

Asian Journal of Pharmaceutical and Clinical Research

Vol. 4, Issue 4, 2011 ISSN - 0974-2441

Research Article

SYNTHESIS, ANTIBACTERIAL, ANTIFUNGAL AND ANTIOXIDANT ACTIVITY STUDIES ON 6-BROMO-2-SUBSTITUTEDPHENYL-1*H*-IMIDAZO [4, 5-b] PYRIDINE

P. LAVANYA, M. SURESH, Y.KOTAIAH, N.HARIKRISHNA, C. VENKATA RAO*

Department of Chemistry, Sri Venkateswara University, Tirupati 517502, India. Email: cvrsvu@gmail.com

Received: 9 Aug 2011, Revised and Accepted: 2 Sep 2011

ABSTRACT

5-Bromopyridine-2,3-diamine **1** underwent facile condensation with various aromatic carboxylic acid derivatives in the presence of Etan's reagent was refluxed and afforded corresponding 6-bromo-2-substitutedphenyl-1*H*-imidazo[4,5-b]pyridine derivatives **3a-j.** Their chemical structures were characterized by using IR, ^{1}H NMR and Mass spectral studies. All the synthesized compounds were screened for anti-microbial and anti-oxidant activities. Most of the compounds showed to potent and significant results when compared to the respective standards.

Keywords: 5-Bromopyridine-2,3-diamine, Aromatic carboxylic acid derivatives; 6-Bromo-2-substitutedphenyl-1*H*-imidazo[4,5-b]pyridine, Antibacterial, Antifungal and antioxidant activity.

INTRODUCTION

For a long time heterocycles have constituted one of the largest areas of research in organic chemistry? Heterocyclic compounds are of particular importance as they are associated with a wide variety of physiological activities with wide variety of heterocyclic systems known today. The nitrogen heterocycles are of great importance as they are present in nucleic acids, vitamins, proteins and other biologically important molecular systems. Among different nitrogen heterocycles, the imidazole and benzimidazole ring systems are very important since several of its derivatives have been found to be medicinally useful. With increase in the incidence of multi drug–resistant to Gram-positive and Gram-negative bacteria it becomes imperative to continuously search for small molecules as anti-infective agents.

Imidazoles and benzimidazoles fit this requirement well since they have demonstrated a diverse set of biological activities that include antibacterial, antiamoebic, antiviral, antifungal[1-4], anthelmintic[5], antiHIV[6], antihistaminic[7], antiulcer[8,9], cardiotonic[10], antihypertensive[11,12] and neuroleptic[13]. Their observed activity depends upon the functional group attached to the moiety. In order to obtain more effective chemotherapeutic agents, a variety of reports have been presented on the synthesis and biological evaluation of new imidazoles and benzimidazoles[14]. The development of resistance to current antibacterial therapy continues to search for more effective agents.

As known not only biochemical similarity of the human cell and fungi forms a handicap for selective activity but also the easily gained resistance is the main problem encountered in developing safe and efficient antifungals. The imidazole antifungals such as clotrimazole, miconazole and ketoconazole showed good optical activity, but were only of limited value for systematic administration. But triazole derivatives are the other major chemical group of antifungal azole derivatives. The triazoles (fluconazole and itraconazole) possess a broad spectrum of antifungal activity and reduced toxicity when compared with the imidazole antifungals[15-17]. Metronidazole and related N-1 substituted 5-nitroimidazoles like ornidazole, secnidazole and tinidazole are widely used in the treatment of diseases caused by protozoa and anaerobic bacteria[18,19].

In view of the above it is worthwhile to prepare imidazole and benzimidazoles derivatives. In view of our ongoing work and inerest in this kind of reaction, our new approach, described in this paper, was to synthesize the imidazopyridines (scheme-I), which were found to have effective biological activities.

MATERIALS AND METHODS

The IR spectra were recorded in KBr discs (υ_{max} in cm^{-1}) on Perkin-Elmer FT-IR spectrophotometer. The ^{1}H-NMR spectra were recorded at 300 M Hz with a Bruker Avance DPX 300 instrument. Mass spectra under electron impact conditions (EI) were recorded at 70 ev ionizing voltage with a VG Prospec instrument and the presented as m/z (% rel int.). Elemental analyses (C,N,H) results were found to be in good agreement with the calculated values. Melting points were determined with Capillaries Thomas Hoover melting point apparatus and are uncorrected. TLC monitored all reactions and purity of the synthesized compounds.

Experimental Work

General Procedure

6-Bromo-2-substituted-1*H*-imidazo[4,5-b]pyridine derivatives (3a-j) were prepared by treating 6-bromo-2,3-diamino-4-pyridine (1) (1mmol), aromatic carboxylic acids (2) 1.1mmol and a mixture of catalyst P_2O_5 and methanesulfonic acid at 100 ^oC for 4h refluxed. Then the contents were neutralized and extracted with ethyl acetate. The solvent was removed under reduced pressure to obtain the compound (3a-j).

3a: 6-Bromo-2-(2,3,4,5-tetraflourophenyl)-1*H*-imidazo[4,5-b]pyridine

Yield 80%, m.p.: 257-258 °C. IR spectrum, υ, cm^{-1} **:** 3421, 3134, 1634, 1574, 1417, 1225, 917. ^{1}H NMR spectrum ($CDCl_3$), δ, ppm **:** 8.17 (s, 1H, H-6'), 8.23 (s, 1H, H-5), 8.50 (s, 1H, H-7), 10.58 (s, 1H, NH). LCMS Mass spectrum, *m/z* **:** 346.4 $(M+1)^+$ and 348.4 $(M+2+H)^+$. Found, % **:** C,41.69; H, 1.18; N, 12.15. $C_{12}H_4BrF_4N_3$. Calculated, % **:** C, 41.65; H, 1.17; N, 12.14.

3b : 6-Bromo-2-pentafluorophenyl-1*H*-imidazo[4,5-b]pyridine

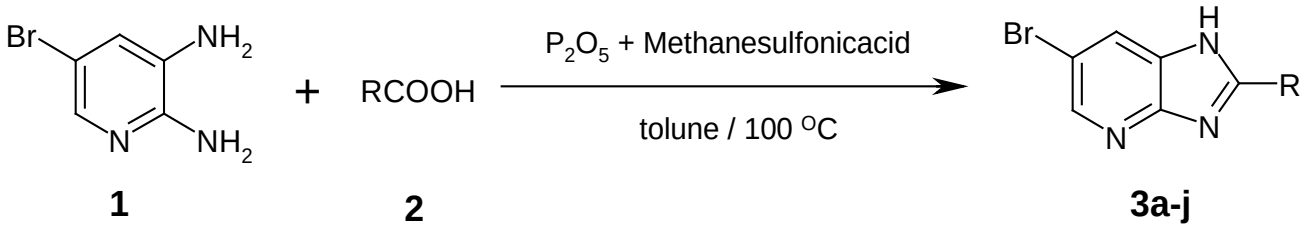
Yield 73%, m.p. **:** 242-244 °C. IR spectrum, υ, cm^{-1} **:** 3436, 3052, 1619, 1573, 1421, 1226, 906. ^{1}H NMR spectrum ($CDCl_3$), δ, ppm **:** 8.35 (s, 1H, H-5), 8.52 (s, 1H, H-7), 10.45 (s, 1H, NH). LCMS Mass spectrum, *m/z* **:** 364 $(M+1)^+$ and 366 $(M+2+H)^+$. Found, % **:** C, 39.60; H, 0.84; N, 11.55. $C_{12}H_3BrF_5N_3$. Calculated, % **:** C, 39.59; H, 0.83; N, 11.54.

3c : 6-Bromo-2-(2-*p*-tolylvinyl)-1*H*-imidazo[4,5-b]pyridine

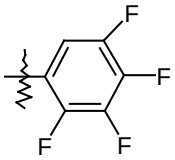
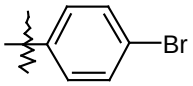
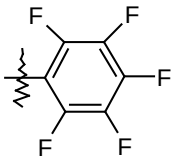
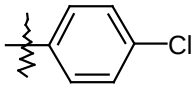
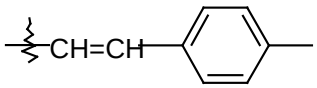
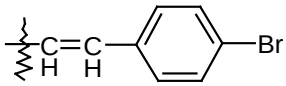
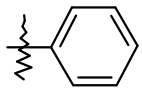
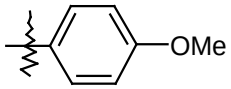
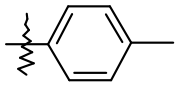
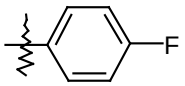
Yield 85%, m.p. **:** 250-252 °C. IR spectrum, υ, cm^{-1} **:** 3394, 3082, 1643, 1563, 1385, 1158, 945. ^{1}H NMR spectrum ($CDCl_3$), δ, ppm **:** 2.21 (s, 3H, CH_3), 7.37 (s, 1H, CH), 7.43 (s, 1H, CH), 7.60 (d, 2H, H-3',5'), 8.0 (d, 2H, H-2',6'), 8.20 (s, 1H, H-5), 8.38 (s, 1H, H-7), 10.50 (s, 1H, NH). LCMS Mass spectrum, *m/z* **:** 314.3 $(M+1)^+$ and 316.3 $(M+2+H)^+$.

Found, %: C, 57.35; H, 3.86; N, 13.38. $C_{15}H_{12}BrN_3$. Calculated, % **:** C, 57.34; H, 3.85; N, 13.37.

Synthetic Scheme



1 **2** **3a-j**

Compound	R	Compound	R
3a		**3f**	
3b		**3g**	
3c		**3h**	
3d		**3i**	
3e		**3j**	

3d : 6-Bromo-2-phenyl-1*H*-imidazo[4,5-b]pyridine : Yield 87%, m.p. **:** 181-182 °C. IR spectrum, υ, cm^{-1} **:** 3448, 3052, 1620, 1545, 1374, 1150, 949. ^1H NMR spectrum ($CDCl_3$), δ, ppm **:** 7.50 (s, 1H, H-4'), 7.61-7.70 (d, 2H, H-3',5'), 8.22-8.30 (d, 2H, H-2',6'), 8.42 (s, 1H, H-5), 8.59 (s, 1H, H-7), 10.50 (s, 1H, NH). LCMS Mass spectrum, *m/z* **:** 274.2 $(M+1)^+$ and 276.2 $(M+2+H)^+$. Found, % **:** C, 52.59; H, 2.95; N, 15.34. $C_{12}H_8BrN_3$. Calculated, % **:** C, 52.58; H, 2.94; N, 15.

3e : 6-Bromo-2-*p*-tolyl-1*H*-imidazo[4,5-b]pyridine : Yield 65%, m.p. **:** 213-214 °C. IR spectrum, υ, cm^{-1} **:** 3428, 3062, 1630, 1560,1369, 1155, 952. ^1H NMR spectrum ($CDCl_3$), δ, ppm **:** 2.15 (s, 3H, CH_3), 7.46 (d, 2H, H-3',5'), 8.16 (d, 2H, H-2',6'), 8.38 (s, 1H, H-5), 8.45 (s, 1H, H-7), 10.55 (s, 1H, NH). LCMS Mass spectrum, m/z **:** 288.1 $(M+1)^+$ and 290.2 $(M+2+H)^+$. Found, % **:** C, 54.20; H, 3.51; N, 14.59. $C_{13}H_{10}BrN_3$. Calculated, % **:** C, 54.19; H, 3.50; N, 14.58.

3f : 6-Bromo-2-(4-bromophenyl)-1*H*-imidazo[4,5-b]pyridine : Yield 80%, m.p. **:** 270-271 °C. IR spectrum, υ, cm^{-1} **:** 3437, 3065, 1610, 1570, 1355, 1162, 955. ^1H NMR spectrum ($CDCl_3$), δ, ppm **:** 7.31 (d, 2H, H-3',5'), 7.59 (d, 2H, H-2',6'), 8.19 (s, 1H, H-5), 8.38 (s, 1H, H-7), 10.58 (s, 1H, NH). LCMS Mass spectrum, *m/z* **:** 353.3 $(M+1)^+$ and 355.1 $(M+2+H)^+$. Found, % **:** C, 40.85; H, 2.01; N, 11.92. $C_{12}H_7Br_2N_3$. Calculated, % **:** C, 40.83; H, 2.00; N, 11.90.

3g : 6-Bromo-2-(4-chlorophenyl)-1*H*-imidazo[4,5-b]pyridine : Yield 80%, m.p. **:** 192-193 °C. IR spectrum, υ, cm^{-1} **:** 3440, 3026, 1635, 1545, 1426, 1236, 923. ^1H NMR spectrum ($CDCl_3$), δ, ppm **:** 7.55 (d, 2H, H-3',5'), 7.86 (d, 2H, H-2',6'), 8.20 (s, 1H, H-5), 8.40 (s, 1H, H-7), 10.45 (s, 1H, NH). LCMS Mass spectrum, *m/z* **:** 308 $(M+1)^+$ and 310 $(M+2+H)^+$. Found, % **:** C, 46.72; H, 2.30; N, 13.63. $C_{12}H_7BrClN_3$. Calculated, % **:** C, 46.71; H, 2.29; N, 13.62.

3h : 6-Bromo-2-[2-(4-bromophenyl)vinyl]-1*H*-imidazo[4,5-b]pyridine : Yield 81%, m.p. **:** 210-211 °C. IR spectrum, υ, cm^{-1} **:** 3445, 3056, 1655, 1563, 1392, 1163, 950. ^1H NMR spectrum ($CDCl_3$), δ, ppm **:** 7.19 (s, 1H, CH), 7.30 (s, 1H,CH), 7.49 (d, 2H, H-3',5'), 8.10 (d, 2H, H-2',6'), 8.20 (s, 1H, H-5), 8.38 (s, 1H, H-7), 10.52 (s, 1H, NH).

LCMS Mass spectrum, *m/z* **:** 379.1 $(M+1)^+$ and 381 $(M+2+H)^+$. Found, % **:** C, 44.37; H, 2.40; N, 11.10. $C_{14}H_9Br_2N_3$. Calculated, % **:** C, 44.36; H, 2.39; N, 11.09.

3i : 6-Bromo-2-(4-methoxyphenyl)-1*H*-imidazo[4,5-b]pyridine : Yield 88%, m.p. **:** 210-211 °C. IR spectrum, υ, cm^{-1} **:** 3439, 3046, 1623, 1578, 1362, 1163, 941. ^1H NMR spectrum ($CDCl_3$), δ, ppm **:** 3.52 (s, 3H, $-OCH_3$), 7.51 (d, 2H, H-3',5'), 8.24 (d, 2H, H-2',6'), 8.35 (s, 1H, H-5), 8.45 (s, 1H, H-7), 10.55 (s, 1H, NH). LCMS Mass spectrum, *m/z***:** 304.1 $(M+1)^+$ and 306.2 $(M+2+H)^+$. Found, % **:** C, 51.35; H, 3.32; N, 6.17. $C_{13}H_{10}BrN_3O$. Calculated **:** C, 51.34; H, 3.31; N, 6.19.

3j : 6-Bromo-2-(4-fluorophenyl)-1*H*-imidazo[4,5-b]pyridine : Yield 83%, m.p. **:** 219-220 °C. IR spectrum, υ, cm^{-1} **:** 3444, 3026, 1610, 1582, 1364, 1132, 945. ^1H NMR spectrum ($CDCl_3$), δ, ppm **:** 7.66 (d, 2H, H-3',5'), 8.14 (d, 2H, H-2',6'), 8.35 (s, 1H, H-5), 8.50 (s, 1H, H-7), 10.52 (s, 1H, NH). LCMS Mass spectrum, *m/z* **:** 292.0 $(M+1)^+$ and 294.1 $(M+2+H)^+$. Found, % **:** C, 49.35; H, 2.43; N, 14.39. $C_{12}H_7BrFN_3$. Calculated, % **:** C, 49.34; H, 2.42; N, 14.38

Table I: Antibacterial activity* of the target compounds 3a-j

Compound	Concentration	Zone of inhibition (mm)			
		Gram-positive bacteria		Gram-negative bacteria	
	(µg)	*S.aureus*	*B.subtilis*	*P.vulgaris*	*K.pneumoniae*
3a	100	17	19	15	16
	200	20	21	18	17
3b	100	17	19	16	18
	200	19	23	17	20
3c	100	31	33	28	30
	200	35	36	31	29
3d	100	15	13	11	14
	200	17	15	15	16
3e	100	11	11	11	12
	200	14	13	14	16
3f	100	30	33	30	31
	200	35	36	34	35
3g	100	27	25	22	24
	200	30	27	28	30
3h	100	31	35	30	31
	200	38	38	34	34
3i	100	30	24	29	24
	200	32	26	30	27
	100	27	28	25	26
3j	200	28	29	27	28
Chloramphenicol	100	35	38	40	42
	200	39	41	44	45

* c = 100 µg / ml; * c = 200 µg / ml.

Biological Study

Antimicrobial Testing

The compound **3a-j** were tested for in vitro antimicrobial activity at two different concentrations 100 and 200µg per disc.

The antibacterial activity was screened against *Staphylococcus aureus*, *Bacillus subtilis* (Gram-positive bacteria) and *Proteus vulgaris, Klebsiella pneumoniae* (Gram-nagative bacteria) on nutrient agar plates at 37 °C for 24 hrs using chloramphenicol as reference during. The compounds were also evaluated for their antifungal activity against *Aspergillus niger* and *Pencillium chrysogenium* using fluconazole as standard drug. Fungi cultures were grown on potato dextrose agar medium (PDA) at 25 °C. The spore suspension was adjusted to 10^6 pores ml^{-1} at an mg ml^{-1} concentration by the Vincent and Vincent method[20].

Antioxidant Testing

The compounds **3a-j** is tested for antioxidant property by nitric oxide and DPPH methods.

Assay for Nitric Oxide (NO) Scavenging Activity Sodium nitroprusside (5µM) in phosphate buffer pH 7.4 was incubated with 100 µM concentration of test compounds dissolved in a suitable solvent (methanol) and tubes were incubated at 25 °C for 120 min. Control experiment was conducted with equal amount of solvent in an identical manner. At intervals 0.5 ml of incubation solution was taken and diluted with 0.5 ml of griess reagent (1% sulfanilamide, 0.1% *N*-naphthylethylenediamine dihydrochloride and 2% *O*-phosphoric acid dissolved in distilled water). The absorbance of the chromophore formed during diazotization of nitrite with sulfanilamide and subsequent *N*-naphthylethylenediamine dihydrochloride was read at 546 nm.

Table II: Antifungal activity* of the target compounds 3a-j

Compound	Concentration	Zone of Inhibition (mm)	
	(µg/ml)	*A.niger*	*P.chrysogenium*
3a	100	23	18
	200	25	24
3b	100	16	25
	200	19	28
3c	100	25	34
	200	39	37
3d	100	14	18
	200	27	21
3e	100	20	19
	200	23	20
3f	100	29	30
	200	35	34
3g	100	31	28
	200	36	31
3h	100	32	33
	200	36	37
3i	100	27	25
	200	29	27
3j	100	32	36
	200	35	38
Fluconazole	100	38	41
	200	42	44

* c = 100 µg / ml; * c = 200 µg / ml.

Table III: Antioxidant activity* of the target compounds 3a-j

Compound	% Inhibition at 100 µM	
	Nitric oxide scavenging activity	DPPH
3a	86.35*	84.74*
3b	43.33	45.12
3c	92.14*	91.69*
3d	28.21	29.55
3e	27.18	26.15
3f	78.24*	81.85*
3g	45.18	49.60
3h	95.32*	95.38*
3i	38.17	41.24
3j	48.28	47.37
Vit. E(5mM)	70±1.22	NT
Curcumin	NT	87.5±3.94
$IC_{50\ for\ 3h}$ (µg/ml)	85.44	77.69

NT= Not tested

Reduction of 1, 1-diphenyl-2-picrylhydrazyl (DPPH) Free Radical (DPPH Method)

The nitrogen centered stable free radical DPPH has often been used to characterize antioxidants. It is reversibly reduced and the odd electron in the DPPH free radical gives a strong absorption maximum at 517 nm, which is purple in color. This property makes it suitable for spectrophotometric studies. A radical scavenging antioxidant reacts with DPPH stable free radical and converts into1,1-diphenyl-2-picrylhydrazine. The resulting decolorization is stoichiometric with respect to the number of electrons captured. The change in the absorbance produced in this reaction has been used to measure antioxidant properties. The solutions of test compounds (100 µM) were added to DPPH (100 µM) in ethanol. The tubes were kept at an ambient temperature for 25 minutes and the absorbance was measured at 517 nm. The difference between the test and the control experiments was taken and expressed as the percentage scavenging of the DPPH radical.

RESULTS AND DISCUSSION

We have prepared 6-bromo-2-substituted phenyl-1*H*-imidazo [4,5-b]pyridine derivatives 3a-j by dehydration of compound 1 with various aromatic carboxylic acid derivatives. Scheme-I

Chemistry

The target 6-bromo-2-(2,3,4,5-tetraflourophenyl)-1*H*-imidazo[4,5-b]pyridine **(3a)** was synthesized by the reaction of 6-bromo-2,3-diamino-4-pyridine (1) and 2,3,4,5-tetrflourobenzoic acid (2a) and a mixture of catalyst P_2O_5 and methanesulfonic acid (Scheme-I). The IR spectrum of (3a) showed absorption bands at 3421 cm^{-1} (NH), 1634 cm^{-1}(C=C) and at 1574 cm^{-1} (C=N). The ^1H NMR spectrum of (3a) displayed signals singlet at δ 8.17 which was assigned to H-6', singlet at δ 8.23 which was assigned to H-5 proton, singlet at δ 8.50 which was assigned to H-7 proton, and singlet at δ 10.58 which was assigned to NH proton. The LCMS mass spectrum of (3a) showed $(M+1)^+$ peak at *m/z* 346.4 and $(M+2+H)^+$ isotopic peak at 348.4 corresponding to its molecular formula $C_{12}H_4BrF_4N_3$

By adopting similar methodology 6-bromo-2-(2-p-tolylvinyl)-1*H*-imidazo[4,5-b]pyridine (3c) was prepared from cinnamic acid (Scheme-I). The IR spectrum of (3c) showed absorption bands at 3394 cm^{-1} (NH), 1643 cm^{-1} (C=C), 1563 cm^{-1} (C=N). The ^1H NMR spectrum of (3c) displayed singlet at δ 2.21 assigned to methyl protons, singlet at δ 7.37 assigned to CH proton, singlet at δ 7.43 assigned to CH proton, doublet at δ 7.60 was assigned to 3',5' protons doublet at δ 8.0 which was assigned to 2',6' protons, singlet at δ 8.20 assigned to H-5 singlet at δ 8.38 assigned to H-7 and singlet at δ 10.50 assigned to NH proton. The LCMS mass spectrum of **(3c)** showed $(M+1)^+$ peak at *m/z* 314.3 and $(M+2+H)^+$ isotopic peak at 316.3 corresponding to its molecular formula $C_{15}H_{12}BrN_3$.

Biological Activity

Imidazopyridines and related derivatives are classes of heterocyclic compounds containing the imidazole nucleus in their structures. Literature survey reveals that a variety of anti microbial agents contains imidazole moiety. In a search for new imidazole systems with potential biological activities, we planned to prepare new 6-bromo-2-substitutedphenyl-1*H*-imidazo[4,5-b]pyridine derivatives. The synthesis of target novel compounds 6-bromo-2-substitutedphenyl-1*H*-imidazo[4,5-b]pyridine derivatives (3a-j) was achieved according to the steps indicated in scheme-1. These reactions are simple, easily carried under normal reaction conditions. We have chosen four bacterial and two fungal strains for microbial studies of these entire imidazole compounds. From this study it is evident that all imidazole compounds showing excellent activity against gram positive bacteria and against gram negative bacteria. Furthermore, the most potent activity was observed in 3a-j against all bacterial and fungal strains when compared to respective standard drugs Chloremphenicol and Fluconozole respectively. The above tested compounds were showed potential free radical scavenging activities like nitric oxide and DPPH to respective standard drugs Vit.E and Curcumin. 3a and 3f posses moderate activity, 3c with good activity. Among the all derivatives 3h has potential activity. A possible explanation for this result is that the biological activity of compounds may be depending on the basic skeleton of molecule as well as on the nature of substituents.

ACKNOWLEDGMENT

The authors are thankful to IICT, Hyderabad for IR, NMR, Mass spectra and Department of Biochemistry, S.V. University, Tirupati for Biological activities.

REFERENCES

1. Bishop B. C, Chelton E. T. J, Jones A. S, Biochem. Pharmacol. 1964; 13:751-754.
2. Bhari N, Shailendra M, Garza G, Cruz-Vega D. E, Garza K, Saleem F. Naqvi, Maura M. R, Azam A, Bioorg. Med. Chem. Lett. 2002; 12: 8-11.
3. Devivar R. V, Kawashima E, Revankar G. R, Breitenbach J, Kreske E, Drach J,Townsend L, J. Med. Chem. 1994; 37: 2942-2949.
4. Sheehan D. J, Hitchcock C. A, Sibley C. M, Clin. Microbiol. Re. 1999;12 : 40-79.
5. Saimo A. G, Cremieux A. C, Hay J. M, Meulemans A, Giovanangeli M. D,Delaitre B, Coulaud F. P, Lancet. 1983 ; 17 : 652-656.
6. Chimirri A, Grasso S, Monforte A. M, Monforte P, Zappala M, Il Farmaco.1991; 46 : 925-933.
7. Benavides J, Schoemaker H, Dana C, Claustre Y, Delahaye M, Prouteau M,Manoury P, Allen J. V, Scatton B, Langer S. Z, Arbilla S, Arzneim.-Forsch.1995; 45: 551-558.
8. Ishihara K, Ichikawa T, Komuro Y, Ohara S, Hotta K, Arzneim.-Forsch. DruRes. 1994; 44: 827-830.
9. Graham D. Y, Mccullough A, Sklar M, Sontag J. S, Roufail W. M, Stone R, Bishop R. H, Gitlin N, Cagliola A. J, Berman R. S, Humphries T, Digestive Diseases and Sciences. 1990; 35: 66-72.
10. Piazzesi G, Morano L, Ruegg J. C, Arzneim. Forsch. Drug Res. 1987; 37: 1141-1143.

11. Wiedemann I, Peil H, Justus H, Adamus S, Brantl V, Lohmann H, Arzneim.-Forsch. Drug Res.1985; 35: 964-969.
12. Kubo K, Kohara Y, Imamiya E, Sugiura Y, Inada Y, Furukawa Y, Nishikawa K, Naka T, J. Med. Chem, 1993; 36: 2182-2195.
13. Janssen P. A. J, Allewijn F. T. N, Arzneim.-Forsch./Drug Res.1968; 18: 279- 282.
14. Nakano H., Inoue T, Kawasaki N, Miyataka H, Matsumoto H, Taguchi T, Inagaki N, Nagai H, Satoh T, Bioorg. Med. Chem.2000; 8: 373-380.
15. Fromtling R. A, Clin. Microbiol. Rev. 1988; 1: 187-208.
16. Odds F. C, Abbott A. B, J. Antimicrob. Chemother. 1984; 14: 105-114.
17. Shadomy S, White S. C, Yu H. P, J. Infect. Dis.1985; 152,:1249-1256.
18. Demir-Erol D, Calıs U, Demirdamar R, Yulug N, Ertan M, J. Pharm. Sci.1995; 4: 462-465.
19. Bhatia S. C and Shanbhag V. D, J. Chromatogr**.** 1984; 305: 325-334.
20. Nielsen C, Pedersen E. B, Bioorganic & Medicinal Chemistry. 2005; 13: 4209-4220.