Int. J. Chem. Sci.: 8(2), 2010, 877-891

SYNTHESIS AND ANTIBACTERIAL ACTIVITY OF 2-SUBSTITUTED BENZIMIDAZOLE-1-CARBODITHIOATE

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ABSTRACT

A series of methyl 2-substituted benzimidazole-1-carbodithioates was successfully synthesized. Benzimidazoles were prepared by condensation of o-phenylenediamine with substituted carboxylic acid and by condensation of substituted benzaldehydes, sodium metabisulphate and o-phenylenediamine. Methyl carbodithioate derivatives of the benzimidazole were prepared by reaction with carbon disulfide. *In vitro* antibacterial activity of the synthesized compounds was analyzed against three gram-positive bacterial species and gram negative microorganism by agar well-diffusion method (Cup plate method).

Key words: Methyl 2-substituted benzimidazole-1-carbodithioates, Benzimidazole, Antibacterial activity, Agar well-diffusion method

INTRODUCTION

On the basis of exhaustive literature review, it is concluded that a little work has been done in field of antibacterial activity of substituted methyl 1*H*-benzimidazole-1-carbodithioate[1-7].

EXPERIMENTAL

All the reactions were monitored using thin layer chromatography (TLC). The melting points of all compounds were determined using open capillary tube melting point apparatus (EIE Instruments, T-0603105) and are uncorrected. The infrared spectra were recorded using KBr as the medium, using JASCO FT-IR-6100 model in the institute. The proton NMR spectra were measured at CSMCRI, Bhavanagar. The mass spectra were recorded at Panjab University, Chandigarh.

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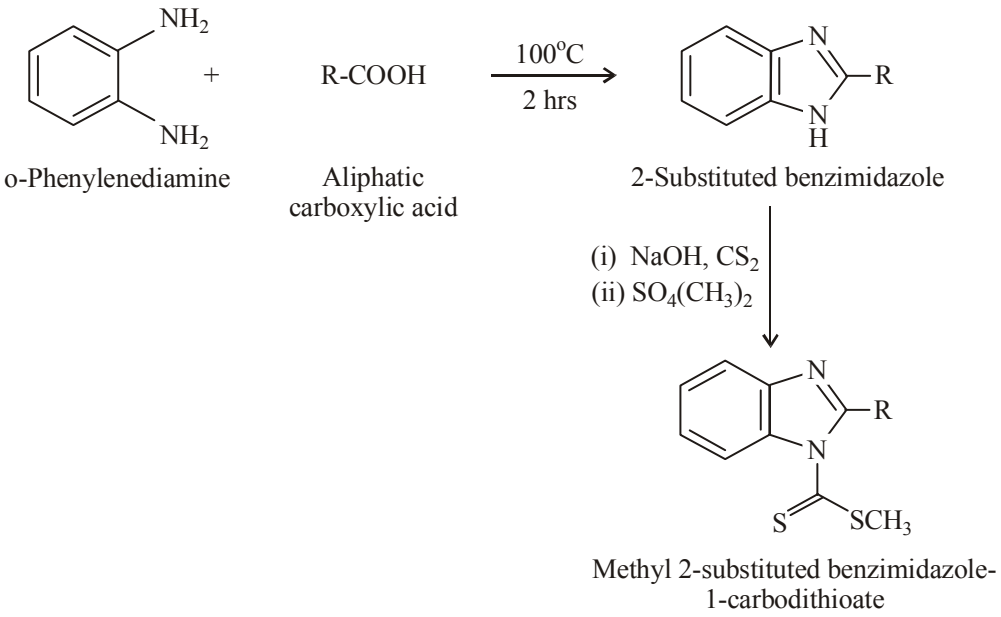
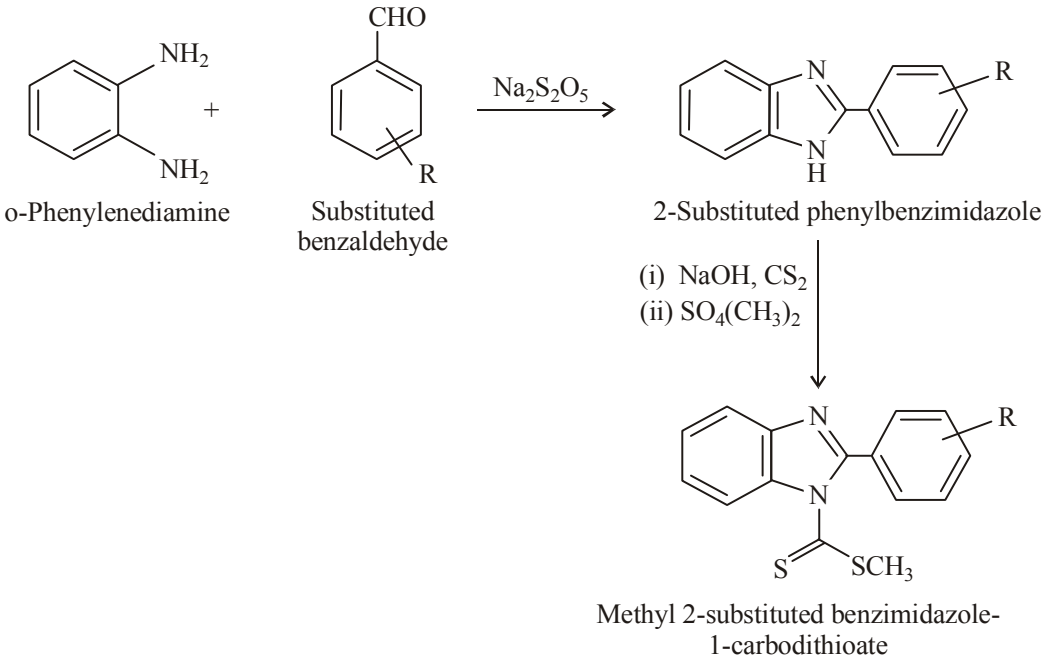


Scheme 1



Scheme 2

Synthesis of benzimidazole

Method 1 (Procedure for compound 1, 2 & 3)

o-Phenylenediamine (6.0 g, 0.055 mole) and substituted carboxylic acid (5.1 g, 0.111 mole) were placed in a round bottom flask. The mixture was heated on a water bath at 100 ºC for 2 hours. The reaction mixture was cooled and concentrated ammonia solution was added slowly dropwise, with constant stirring, until the mixture was just alkaline to litmus. Crude benzimidazole was filtered off at vacuum pump, washed with ice-cold water, and recrystallized from 10 % v/v aqueous ethanol. Solvent system used for TLC was n-hexane : ethyl acetate (7 : 3).

Method 2 (Procedure for compound 4 to 13)

Benzaldehyde (5.0 g, 0.047 mole) was added to methanol (50.0 mL) in a beaker and stirred for few minutes. Saturated solution of sodium metabisulfite (10 mL) was added portion wise into this mixture with constant stirring. The mixture was stirred vigorously and more methanol was added. The mixture was kept in a refrigerator over night. The resulting precipitate was filtered and dried properly (yield more than 90-95%).

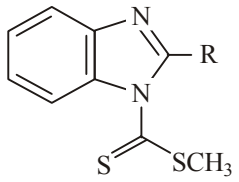
o-Phenylenediamine (4.1 g, 0.038 mole) was dissolved in DMF (40.0 mL) and benzaldehyde adduct (8.0 g, 0.038 mole) was added into it. The mixture was heated under reflux for 4 hours. The reaction mixture was cooled and poured into cold water. The resulting solid was filtered, dried and recrystallized from ethanol. Solvent system used for TLC was n-hexane : ethyl acetate (7 : 3).

Synthesis of methyl carbodithioate derivatives of benzimidazole

Benzimidazole was dissolved in DMSO in 100 mL round bottom flask and this solution was stirred on a magnetic stirrer for 5-10 minutes at room temperature. Carbon disulfide and aqueous sodium hydroxide (20 mole solution) was added dropwise simultaneously over 20-30 minutes and it was further stirred for 1 hour at room temperature. After an hour, dimethyl sulphate was added dropwise to this reaction mixture with stirring at 5-10 ºC maintained using ice-bath. It was further stirred for 2-3 hours at 5-10 ºC and then poured onto crushed ice with constant stirring. The solid obtained was filtered by vacuum filtration, dried and recrystallized from ethanol.

The structures of novel synthesized compounds of benzimidazole series were elucidated by IR, ^{1}H NMR, and Mass spectroscopic data. The summary of physical data and spectral analysis data of the compound **1** to **13** are shown in Table 2 and 3, respectively.

Table 1: Structure and physical data of the synthesized compounds



Compd.	R	Molecular formula	MASS	M.P. (ºC)	R_f
1	-H	$C_9H_8N_2S_2$	208.29	58	0.86
2	-CH_3	$C_{10}H_{10}N_2S_2$	222.41	266	0.76
3	-CH_2-Ph	$C_{16}H_{14}N_2S_2$	298.40	159	0.70
4	-Ph	$C_{15}H_{12}N_2S_2$	284.32	260	0.59
5	-4'-N(CH_3)$_2$-Ph	$C_{17}H_{17}N_3S_2$	327.44	270	0.52
6	- 3'-OCH_3-Ph	$C_{16}H_{14}N_2OS_2$	314.4	216	0.79
7	-3',4'-(OCH_3)$_2$-Ph	$C_{17}H_{16}N_2O_2S_2$	344.42	205	0.67
8	-2'-Cl-Ph	$C_{15}H_{11}N_2S_2Cl$	318.82	212	0.4
9	-3'-Cl-Ph	$C_{15}H_{11}N_2S_2Cl$	318.82	220	0.46
10	-4'-Cl-Ph	$C_{15}H_{11}N_2S_2Cl$	318.82	257	0.81
11	-2'-NO_2-Ph	$C_{15}H_{11}N_3O_2S_2$	329.37	263	0.62
12	-3'-NO_2-Ph	$C_{15}H_{11}N_3O_2S_2$	329.37	167	0.73
13	-4'-NO_2-Ph	$C_{15}H_{11}N_3O_2S_2$	329.37	257	0.55

Table 2: Spectroscopic data of the synthesized compounds

Compd.	IR Data
1	IR (KBr, cm^{-1}): 3092 (aromatic C-H stretching), 1603 & 1510 (C=C aromatic stretching), 1365 (C-H bending of methyl carbodithioate group), 1287 (C-N stretching), 1054 (C=S stretching) and 766 (C-S stretching)
2	3193, 1579 & 1489, 1372, 1231, 1013 and 682

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Compds	IR Data
3	3079, 3018, 1588 & 1488, 1363, 1223, 1012, 725 and 694
4	3047, 1590 & 1495, 1374, 1227, 1006, 764 and 658
5	3075, 1614 & 1504 , 1398, 1225, 1011, 804 and 731
6	3060, 1592 & 1481, 1322, 1288, 1233, 1110, 809 and 787
7	3018, 1608 & 1504, 1387, 1256, 1228, 1027 and 767 Mass (ESI-MS) (*m/z* [M+1], %): 345.17, 50 % ^{1}H NMR (DMSO-d_6, δ, ppm): 3.84 (s, 3*H*, OCH_3 at C_4), 3.89 (s, 3*H*, OCH_3 at C_3), 7.12-7.78 (m, 7*H*, aromatic protons), 12.75 (s, 3*H*, SCH_3).
8	3047, 1591 & 1489, 1401, 1232, 1035, 810, 742 and 732
9	3047, 1604 & 1488, 1412, 1228, 1114, 796, 798 and 677
10	3052, 1603 & 1449, 1365, 1226 , 1040 , 829, 745 and 729
11	3025, 1608 & 1504, 1554, 1368, 1349, 1241, 1016, 808 and 756
12	3059, 1624 & 1501, 1522, 1387, 1349, 1228, 1071, 791 and 706
13	2999, 1604 & 1524, 1517 1399, 1339, 1227, 1011, 852 and 709

RESULTS AND DISCUSSION

In vitro antibacterial activity of the synthesized compounds was analyzed against three gram-positive bacterial species [*Staphylococcus aureus* (MTCC 737), *Enterococcus faecalis* (MTCC 439) and *Bacillus cereus* (MTCC 430)] and three gram-negative bacterial species [*Escherichia coli* (MTCC 1687), *Pseudomonas aeruginosa* (MTCC 2642) and *Klebsiella pneumoniae* (MTCC 109)] by agar well-diffusion method (Cup plate method).[8,9] Four serial dilutions yielding concentrations of 700, 500, 300 and 100 mg per well for the all 13 synthesized compounds were used. Ciprofloxacin was used as a standard reference. DMF was used as a control, which did not exhibit any inhibition. The petri dishes were incubated at 37 ± 1 °C for 24 hrs. The diameter of zone of inhibition produced by each compound was measured in millimeter using Antibiotic Zone Reader (Hally Instruments) and the results are presented in Table 3 and in Figs. 1 to 6. The antibacterial activity was determined in triplicate[10-14].

Table 3: Zone of inhibition of the synthesized compounds against selected bacterium species

CMPND	Zone of inhibition (mm) of the Synthesized compounds											
	Escherichia coli				*Enterococcus faecalis*				*Pseudomonas aeruginosa*			
	700 µg/well	**500 µg/well**	**300 µg/well**	**100 Mg/well**	**700 µg/well**	**500 µg/well**	**300 µg/well**	**100 µg/well**	**700 µg/well**	**500 µg/well**	**300 µg/well**	**100 µg/well**
STD	44.2 ± 0.15	44.5 ± 0.10	43.0 ± 0.25	42.4 ± 0.15	35.1 ± 0.10	30.2 ± 0.15	29.8 ± 0.10	28.3 ± 0.21	44.4 ± 0.21	44.2 ± 0.21	43.9 ± 0.10	43.2 ± 0.15
1	10.8 ± 0.15	10.2 ± 0.15	8.1 ± 0.21	4.1 ± 0.21	22.8 ± 0.10	22.6 ± 0.10	18.8 ± 1.24	14.4 ± 0.32	22.3 ± 0.21	20.0 ± 0.38	10.3 ± 0.15	4.1 ± 0.15
2	3.2 ± 0.15	2.1 ± 0.21	0.0 ± 0.00	0.0 ± 0.00	10.2 ± 0.15	8.4 ± 0.10	8.1 ± 0.15	4.5 ± 0.10	2.7 ± 0.17	2.2 ± 0.15	0.0 ± 0.00	0.0 ± 0.00
3	6.0 ± 0.06	5.7 ± 0.15	5.1 ± 0.10	1.9 ± 0.06	8.4 ± 0.15	6.7 ± 0.17	6.3 ± 0.10	4.1 ± 0.12	1.3 ± 0.10	1.1 ± 0.23	0.0 ± 0.00	0.0 ± 0.00
4	6.4 ± 0.15	6.3 ± 0.10	6.0 ± 0.15	4.2 ± 0.15	6.8 ± 0.06	6.2 ± 0.20	5.7 ± 0.10	4.1 ± 0.12	3.4 ± 0.15	1.2 ± 0.15	0.0 ± 0.00	0.0 ± 0.00
5	6.0 ± 0.25	5.2 ± 0.06	4.8 ± 0.10	4.3 ± 0.31 ±	7.7 ± 0.06	6.5 ± 0.10	6.2 ± 0.06	5.5 ± 0.10	5.8 ± 0.06	4.2 ± 0.21	2.1 ± 0.10	0.0 ± 0.00
6	4.8 ± 0.15	4.0 ± 0.15	3.4 ± 0.20	2.8 ± 0.20	6.5 ± 0.10	6.1 ± 0.10	4.2 ± 0.00	4.0 ± 0.00	4.4 ± 0.15	4.3 ± 0.17	2.2 ± 0.21	0.0 ± 0.00

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CMPND	Zone of inhibition (mm) of the synthesized compounds											
	Escherichia coli				*Enterococcus faecalis*				*Pseudomonas aeruginosa*			
	700 μg/well	**500 μg/well**	**300 μg/well**	**100 μg/well**	**700 μg/well**	**500 μg/well**	**300 μg/well**	**100 μg/well**	**700 μg/well**	**500 μg/well**	**300 μg/well**	**100 μg/well**
7	8.8 ± 0.15	8.2 ± 0.15	6.0 ± 0.25	2.1 ± 0.15	8.2 ± 0.20	6.2 ± 0.15	5.6 ± 0.31	4.2 ± 0.15	6.2 ± 0.26	4.8 ± 0.06	3.9 ± 0.15	2.2 ± 0.26
8	7.2 ± 0.10	6.3 ± 0.15	6.1 ± 0.21	0.0 ± 0.00	4.5 ± 0.10	4.2 ± 0.15	0.0 ± 0.00	0.0 ± 0.00	8.2 ± 0.25	4.6 ± 0.25	2.2 ± 0.25	2.2 ± 0.21
9	6.6 ± 0.21	6.1 ± 0.10	6.1 ± 0.10	4.2 ± 0.20	6.2 ± 0.21	4.9 ± 0.15	4.8 ± 0.10	17.3 ± 0.49	4.8 ± 0.10	4.1 ± 0.15	1.1 ± 0.12	0.0 0.00
10	4.7 ± 0.12	4.0 ± 0.15	3.7 ± 0.40	0.0 ± 0.00	1.6 ± 0.15	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	4.3 ± 0.25	1.7 ± 0.10	1.1 ± 0.15	0.0 ± 0.00
11	5.2 ± 0.15	4.5 ± 0.12	2.2 ± 0.15	0.0 ± 0.00	6.5 ± 0.10	6.3 ± 0.15	4.7 ± 0.10	4.2 ± 0.15	6.4 ± 0.15	4.1 ± 0.15	2.2 ± 0.29	0.0 ± 0.00
12	3.0 ± 0.15	2.8 ± 0.15	2.4 ± 0.21	0.0 ± 0.00	8.7 ± 0.10	8.4 ± 0.15	5.6 ± 0.06	5.4 ± 0.15	4.4 ± 0.25	4.2 ± 0.15	2.4 ± 0.15	0.0 ± 0.00
13	4.6 ± 0.15	4.2 ± 0.15	4.0 ± 0.20	2.8 ± 0.15	0.4 ± 0.21	9.9 ± 0.21	9.9 ± 0.06	9.5 ± 0.25	2.9 ± 0.10	1.0 ± 0.10	0.0 ± 0.00	0.0 ± 0.00

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CMPND	Zone of inhibition (mm) of the synthesized compounds											
	Staphylococcus aureus				*Klebsiella pneumoniae*				*Bacillus cereus*			
	700 µg/well	500 µg/well	300 µg/well	100 µg/well	700 µg/well	500 µg/well	300 µg/well	100 µg/well	700 µg/well	500 µg/well	300 µg/well	100 µg/well
STD	40.2 ± 0.15*	38.5 ± 0.12	36.7 ± 0.15	36. ± 0.10	33.2 ± 0.06*	32.6 ± 0.15	32.1 ± 0.10	32.1 ± 0.15	41.4 ± 0.15*	40.2 ± 0.10	39.9 ± 0.21	38.5 0.12
1	24.4 ± 0.15	20.3 ± 0.15	18.7 ± 0.10	18. ± 0.00	24.8 ± 0.15	24.3 ± 0.12	24.4 ± 0.15	14.2 ± 0.15	24.7 ± 0.12	24.3 ± 0.12	20.2 ± 0.15	14.2 0.20
2	4.2 ± 0.25	3.8 ± 0.15	2.5 ± 0.20	0.0 ± 0.00	4.6 ± 0.21	4.5 ± 0.17	2.1 ± 0.15	2.1 ± 0.10	4.8 ± 0.20	4.4 ± 0.15	4.1 ± 0.10	0.0 0.00
3	8.3 ± 0.10	6.7 ± 0.10	5.3 ± 0.10	2.7 ± 0.10	2.4 ± 0.15	1.7 ± 0.36	1.0 ± 0.15	0.0 ± 0.00	4.6 ± 0.10	4.1 ± 0.10	2.0 ± 0.15	0.0 0.00
4	6.7 ± 0.15	6.3 ± 0.12	6.1 ± 0.10	2.5 ± 0.15	10.2 ± 0.15	8.7 ± 0.15	8.1 ± 0.12	4.1 ± 0.31	8.1 ± 0.12	6.4 ± 0.15	6.1 ± 0.51	2.7 0.10
5	8.3 ± 0.15	6.4 ± 0.25	4.2 ± 0.06	3.8 ± 0.15	4.7 ± 0.21	4.2 ± 0.00	4.1 ± 0.15	3.8 ± 0.10	8.1 ± 0.10	6.0 ± 0.25	5.2 ± 0.06	4.2 0.15
6	8.5 ± 0.10	8.1 ± 0.17	8.0 ± 0.20	4.1 ± 0.06	6.1 ± 0.10	4.5 ± 0.25	4.3 ± 0.10	4.1 ± 0.15	8.6 ± 0.15	8.0 ± 0.15	6.2 ± 0.15	0.0 0.00

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CMPND	Zone of inhibition (mm) of the synthesized compounds											
	Staphylococcus aureus				*Klebsiella pneumoniae*				*Bacillus cereus*			
	700 µg/well	500 µg/well	300 µg/well	100 µg/well	700 µg/well	500 µg/well	300 µg/well	100 µg/well	700 µg/well	500 µg/well	300 µg/well	100 µg/well
7	4.7 ± 0.15	4.2 ± 0.15	3.3 ± 0.10	0.0 ± 0.00	.Ω± 0.25	1.7 ± 0.10	1.1 ± 0.10	0.0 ± 0.00	8.0 ± 0.25	4.1 ± 0.15	2.3 ± 0.10	0.0 ± 0.00
8	2.5 ± 0.15	2.2 ± 0.20	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	2.2 ± 0.21	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
9	2.3 ± 0.15	1.6 ± 0.21	0.0 ± 0.00	0.0 ± 0.00	4.6 ± 0.15	4.0 ± 0.15	2.9 ± 0.10	2.1 ± 0.10	4.5 ± 0.21	4.5 ± 0.10	2.1 ± 0.10	2.1 ± 0.10
10	4.6 ± 0.06	4. ± 0.15	2.2 ± 0.20	2.2 ± 0.15	1.1 ± 0.06	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	2.1 ± 0.10	1.2 ± 0.15	0.0 ± 0.00	0.0 ± 0.00
11	6.4 ± 0.15	6.1 ± 0.06	4.2 ± 0.21	2.1 ± 0.10	8.7 ± 0.15	6.2 ± 0.06	4.1 ± 0.10	2.1 ± 0.06	9.2 ± 0.15	8.3 ± 0.06	4.1 ± 0.15	0.0 ± 0.00
12	4.7 ± 0.17	4.6 ± 0.10	4.2 ± 0.15	0.0 ± 0.00	8.6 ± 0.10	8.1 ± 0.12	7.7 ± 0.21	4.0 ± 0.25	4.7 ± 0.10	4.1 ± 0.10	3.7 ± 0.17	0.0 ± 0.00
13	6.8 ± 0.15	6.2 ± 0.15	4.0 ± 0.06	3.7 ± 0.21	7.2 ± 0.06	6.9 ± 0.10	6.3 ± 0.21	6.2 ± 0.10	8.6 ± 0.21	8.2 ± 0.21	6.1 ± 0.10	6.1 ± 0.17

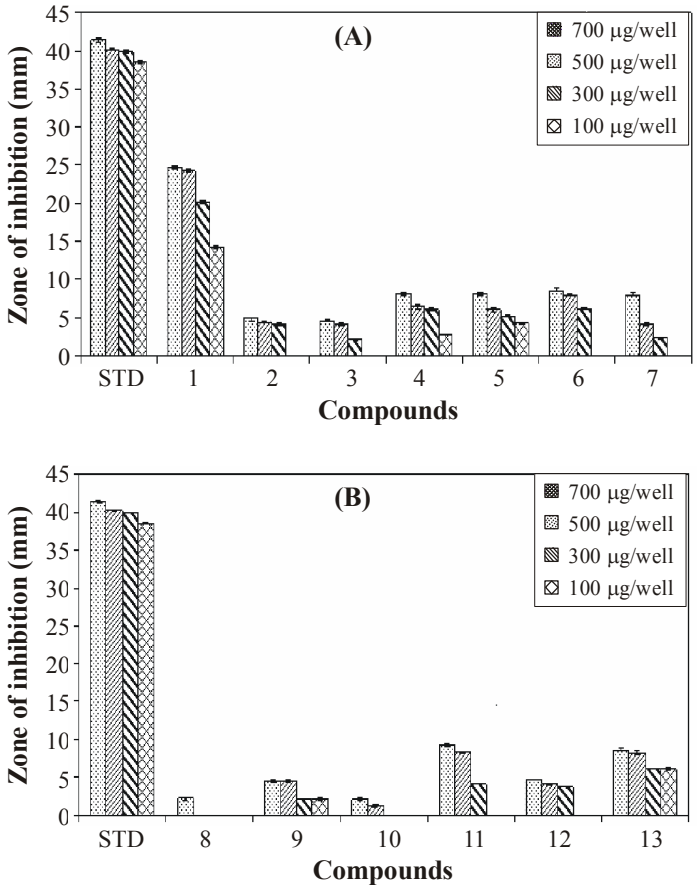
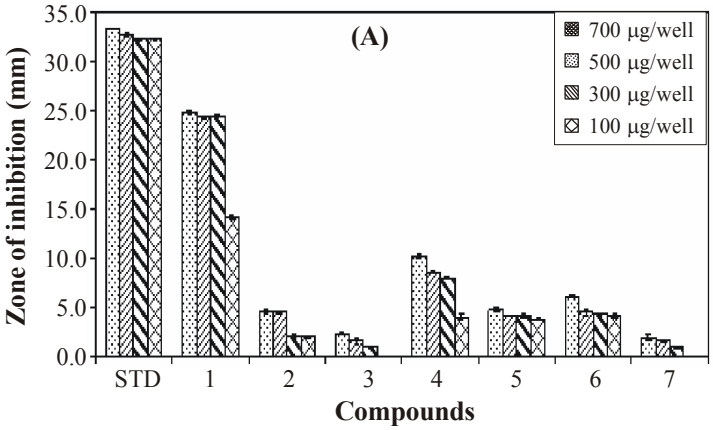


Fig. 1: Zone of inhibition of the synthesized compounds against *B. cereus*



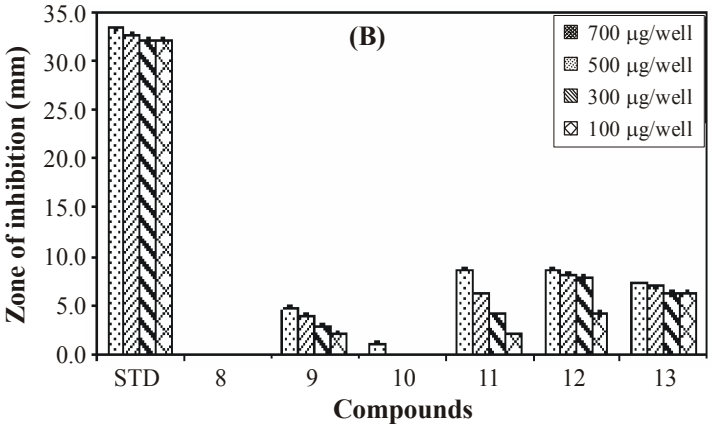


Fig. 2: Zone of inhibition of the synthesized compounds against *K. pneumoniae*

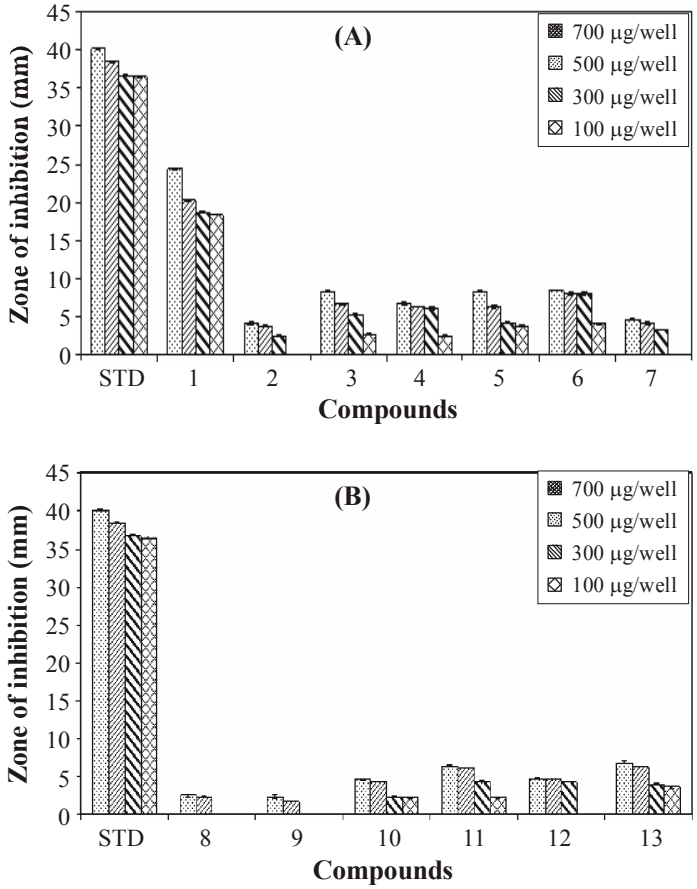


Fig. 3: Zone of inhibition of the synthesized compounds against *S. aureus*

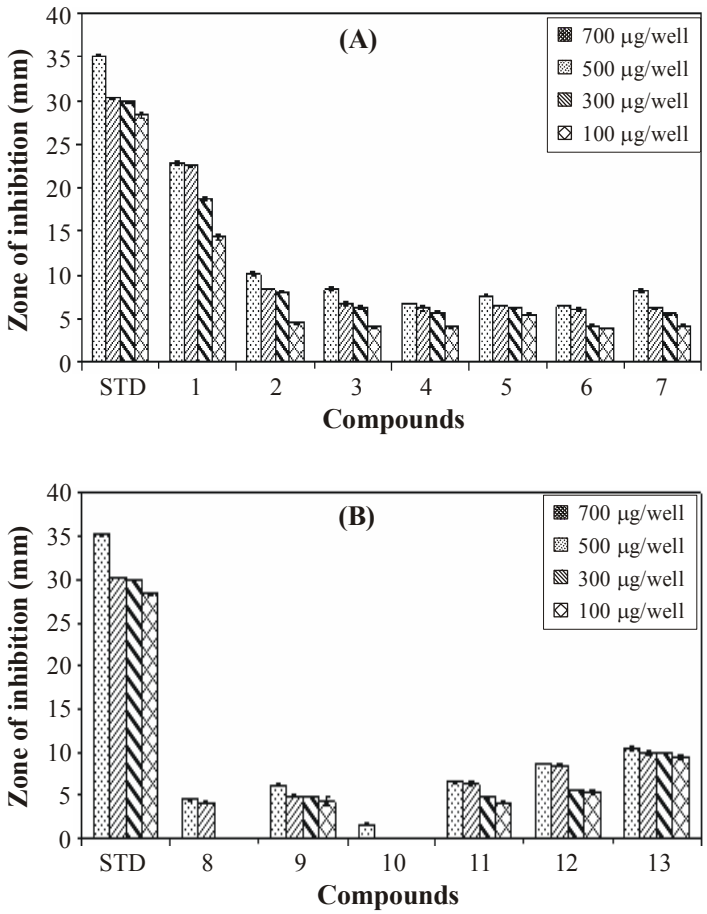
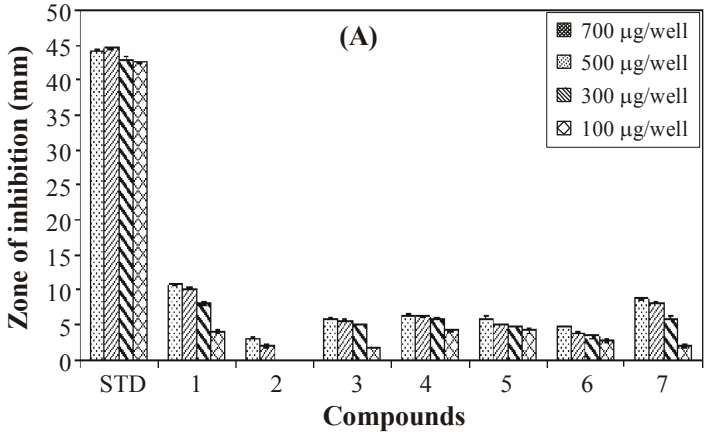


Fig. 4: Zone of inhibition of the synthesized compounds against *E. faecalis*



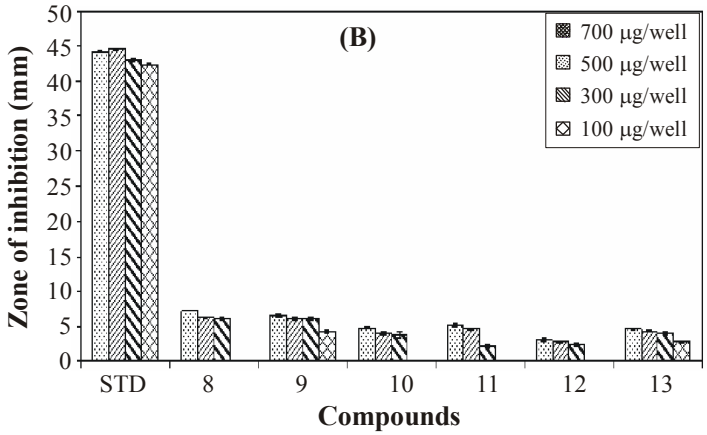


Fig. 5: Zone of inhibition of the synthesized compounds against *E. coli*

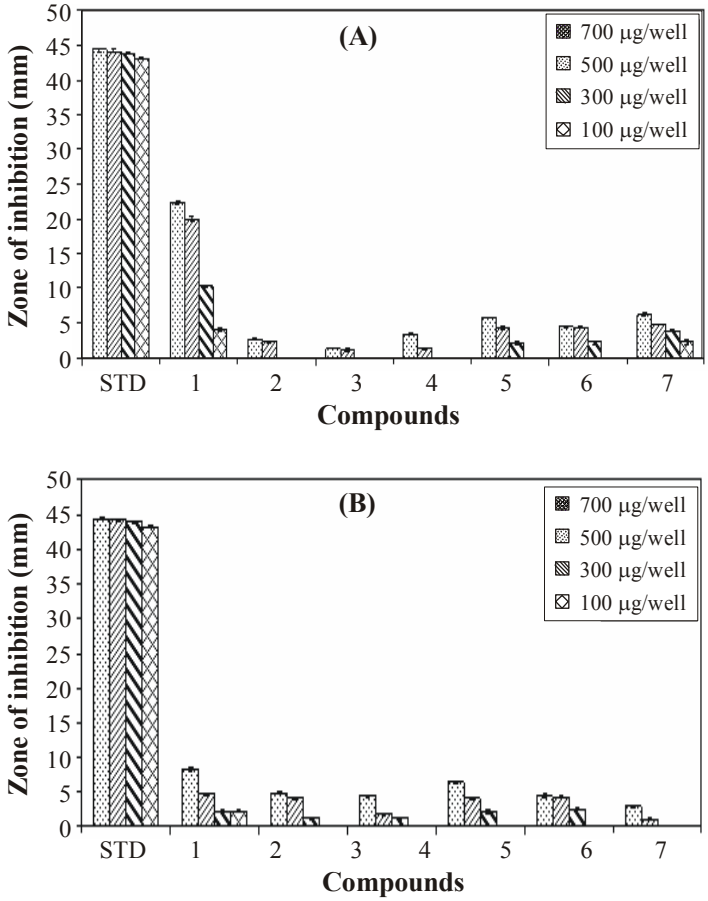


Fig. 6: Zone of inhibition of the synthesized compounds against *P. aeruginosa*

In the present study, the results revealed that most of the synthesized compounds exhibited comparable inhibitory activity to reference standard drug ciprofloxacin. Methyl 1*H*-benzimidazole-1-carbodithioate **(1)** showed more pronounced antibacterial activity than other compounds of this benzimidazole series, with better inhibitory activity against both gram-positive and gram-negative bacteria. Methyl 2-(4′-nitrophenyl)-1*H*-benzimidazole-1-carbodithioate **(13)** exhibited remarkable inhibitory activity against all bacteria except *Ps. aeruginosa* and *E. coli.* Other compounds except methyl 2-methyl-1*H*-benzimidazole-1-carbodithioate **(2)**, methyl 2-(2′-chlorophenyl)-1*H*-benzimidazole-1-carbodithioate **(8)** and methyl 2-(4′-chlorophenyl)-1*H*-benzimidazole-1-carbodithioate **(10)** exhibited average to moderate activity against all test bacteria.

REFERENCES

1. C. Kuş, Turk J. Chem., **26**, 559 (2002)**.**
2. M. Hasan, F. Shaheen and U. Yunus, Turk. J. Chem., **24**, 730 (2000).
3. R. Kumar and Y. C. Joshi, E-J. Chem., **4(4**), 606 (2007).
4. L. M. Yagupolskii, D. V. Fedyuk, K. I Petko, V. I. Troitskaya and V. V Rudyuk, J. Fluorine Chem., **106**, 181 (2000).
5. A. K. Gülgün and A. Nurten, Turk. J. Chem., **30**, 223 (2006).
6. H. Xianging, M. Huiqiang and W. Yulu, Arkivoc., **8**, 150 (2007).
7. V. Alagarsamy, V. R. Solomon, R. Meena, K. V. Ramaseshu, K. Thirumurugan and S. Murugesan, J. Med. Chem. **, 3**, 67 (2007).
8. A. Manoharan, R. Pai, V. Shanhar, T. Thomas and M. K. Lalitha, Indian J. Med. Res., **117**, 81 (2003).
9. C. Valgas, S. Machado de Souza and E. F. A. Smânia, Braz. J. Microbiol., **38**, 369 (2007).
10. J. Jayabharath, V. Thanikachalam, A. Thangami and M. Padmavathy, Synthesis, **16**, 266 (2007).
11. S. Alam, Acta Chim. Slov., **51**, 447 (2004).
12. F. M. Morad, M. M. Ajaily and S. B. Gweirif, J. Sci. Appl., **1(1),** 72 (2007).
13. S. K. Sahu, S. K. Mishra, R. K. Mohanta, S. P. Pattanayak and C. S. Panda, Indian J. Pharm. Sci. **68(3)**, 377 (2006).

14. G. Suleyman, T. Metin, O. Hasan and S. Hasan, Turk. J. Chem., **29**, 199 (2005).

Revised : 08.12.2009 *Accepted : 12.12.2009*