Heterocyclic Letters
Vol. 8| No.3|631-639|May-July|2018
ISSN: (print) 2231–3087 / (online) 2230-9632
CODEN: HLEEAI
http://heteroletters.org

SYNTHESIS AND ANTIMICROBIAL SCREENING OF NOVEL 4-(1*H*-BENZO[*d*] IMIDAZOL-2-YL)-8,9-DIARYL-1,6-DIOXA-4,7,9-TRIAZASPIRO[4,5]DEC-7-EN-3-ONES

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Abstract

The synthesis of novel 4-(1*H*-benzo[*d*]imidazol-2-yl)8,9-diaryl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-ones **(4)** were achieved by the reaction of 2-aminobenzimidazole **(1)** with chloroacetylchloride, followed by cyclization with arylisocyanates, then cycloaddition with benzonitrile oxides. All the newly synthesized compounds **2-4** were characterized on the basis of IR, ^{1}H NMR, ^{13}C NMR and mass spectral data and screened for their antimicrobial activity.

Keywords: Benzimidazolyl 1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-ones, cyclization, cycloaddition, antibacterial activity, antifungal activity.

Introduction

The increasing incidence of bacterial and fungal resistance to a large number of antimicrobial agents has prompted studies on the development of new potential antimicrobial compounds. The molecular manipulation of promising lead compounds is still a major line of approach to develop new drugs. It involves efforts to combine separate pharmacophoric groups of similar activity in to one compound, thus making changes in the biological activity. So, the discovery of novel and potent antimicrobial agents is the best way to overcome microbial resistance and develop effective therapies[i].

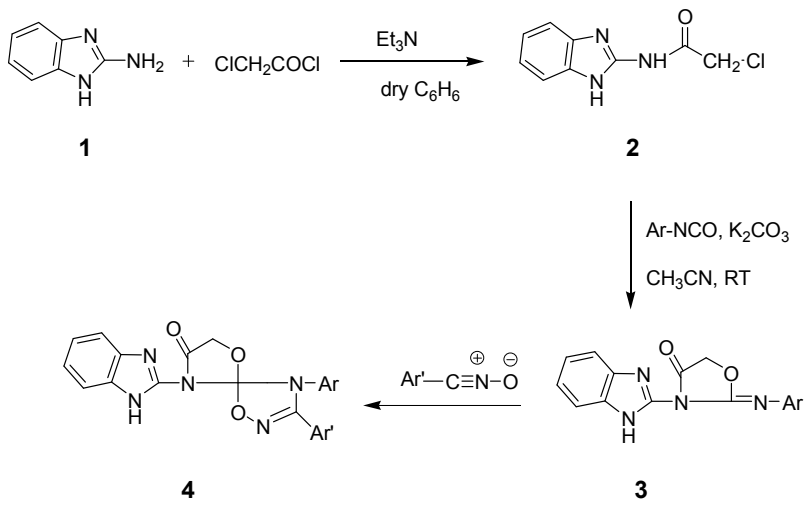
Benzimidazole is an important nucleus that has been extensively used in medicinal chemistry. They are known to exhibit anti-inflammatory[ii], antibiotic[iii], antihelminthic[iv], anticancer[v] and antiviral[vi] activities. Spirooxadiazolines have attracted attention because of their enhanced biological activity. Several of these derivatives are known to possess antiproliferative[vii], antimycobacterial[viii], anticancer[ix,x], anti-inflammatory[xi] and antimicrobial activity[xii]. Realizing the importance of the benzimidazole and 1,2,4-oxadiazolines biodynamic heteryl nuclei, and in continuation of our interest in designing the synthesis of biologically active nitrogen and oxygen heterocycles linked to benzimidazole, it was thought worthwhile to undertake the synthesis of novel spirooxadiazolines containing benzimidazole

moiety to evaluate their antimicrobial activity. As a sequel to our project on the synthesis of benzimidazolyl derivatives with potential activities[xiii,xiv], we, herein, wish to report the synthesis of novel benzimidazolyl 1,6-dioxa-4,7,9-triazaspiro[4,5]-dec-7-ene-3-ones and their antimicrobial activity.

Results and Discussion

The reaction of 2-aminobenzimidazole **(1)** with chloroacetyl chloride in the presence of triethylamine in dry benzene furnished N-(1*H*-benzo[*d*]imidazol-2-yl)-2-chloroacetamide **(2).** Compound **(2)** on treatment with arylisocyanates in the presence of K_2CO_3 in CH_3CN afforded the corresponding 3-(1*H*-benzo[*d*]imidazol-2-yl)-2-(arylimino)oxazolidin-4-ones **(3).** The mechanism involves the addition of amide derivative to isocyanate in the presence of a base and subsequent cyclization takes place by nucleophilic substitution of chlorine by the oxygen atom of isocyanate. This mechanism is in agreement with earlier literature observation[xv]. Cycloaddition of **3** with benzonitrile oxides, generated *in situ* from benzhydroxamoyl chloride in the presence of triethyl amine, at ice-cold temperature furnished the novel 4-(1*H*-benzo[*d*]imidazol-2-yl)-8,9-diaryl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-ones **(4)** by the 1,3-dipolar cycloaddition. The other regioisomer could not be obtained in the reaction. This selectivity in cycloaddition leading to the formation of only one of the possible regioisomers may be due to the polarization of the imine double bond of **(3)** towards nitrogen, making imine carbon positively changed. So, the oxygen of benzonitrile oxide attacks on this carbon giving only product **(4).** The structure of the products **2-4** have been established on the basis of IR, ^{1}H NMR, ^{13}C NMR and MS spectral data. **(Scheme-I).**

Compound **2** displayed characteristic absorption bands in IR spectrum around 1685, 3356 and 3389 cm^{-1} due to amide carbonyl, amide NH and benzimidazole NH functional groups respectively. ^{1}H NMR spectrum of **(2)** exhibited a sharp singlet at δ 4.49 and a broad singlet at δ 8.30 due to methylene and CONH protons respectively. Mass spectrum of **(2)** exhibited molecular ion $[M+H]^+$ peak at *m/z* 210. The ^{1}H NMR spectrum of **(3)** displayed a prominent singlet at δ 4.22 due to newly formed 1,3-oxazalan-4-one.The mass spectrum of **(3)** confirmed the structure by displaying the molecular ion $[M+H]^+$ peak at *m/z* 293. The ^{1}H NMR spectrum of **(4)** shown a sharp singlet at δ 4.10 due to methylene protons. The mass spectrum of **(4)** confirmed the cycloaddition reaction by exhibiting the molecular ion $[M+H]^+$ peak at *m/z* 412. The ^{13}C NMR spectra of **(3)** and **(4)** are in agreement with the proposed structures.



	3		4	
	Ar		**Ar**	**Ar'**
a,	C_6H_5	**a,**	C_6H_5	C_6H_5
b,	4-$OCH_3C_6H_4$	**b,**	4-$OCH_3C_6H_4$	C_6H_5
c,	4-ClC_6H_4	**c,**	4-ClC_6H_4	C_6H_5
d,	4-$CH_3C_6H_4$	**d,**	4-$CH_3C_6H_4$	C_6H_5
e,	4-BrC_6H_4	**e,**	C_6H_5	4-ClC_6H_4
f,	4-$NO_2C_6H_4$	**f,**	C_6H_5	4-$OCH_3C_6H_4$
g,	4-$N(CH_3)_2C_6H_4$	**g,**	4-BrC_6H_4	C_6H_5
h,	2,6-$Cl_2C_6H_3$	**h,**	C_6H_5	4-$NO_2C_6H_4$

Scheme I

Antibacterial activity

The newly synthesized 4-(1*H*-benzo[*d*]imidazol-2-yl)-8,9-diphenyl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-ones (**4a-h)**, were evaluated for their *in vitro* antibacterial activity against Gram-positive bacteria *viz*., *Bacillus subtilis* (MTCC 441), *Bacillus sphaericus* (MTCC 511) and *Staphylococcus aureus* (MTCC 96) and Gram-negative bacteria *viz*., *Pseudomonas aeruginosa* (MTCC 741), *Klobsinella aerogenes* (MTCC 39) and *Chromobacterium violaceum* (MTCC 2656) at 100 µg/ml concentration. The *in vitro* antibacterial activity of the tested compounds was assessed by minimum inhibitory concentration (MIC) using broth dilution method [xvi]. *Ciprofloxacin* was used as standard drug for comparison.

The antibacterial activity results showed that the compounds **4a-h** displayed a better activity and were more active than standard drug *Ciprofloxacin* (**Table 1**). The activity was expressed in terms of minimum inhibitory concentration (MIC). The compounds **4b** and **4d** are highly active, because the activity is considerably affected by the presence of methyl and methoxy groups as substituents on benzene ring, besides the presence of basic skeleton. Compounds **4c, 4e and 4g** carrying chloro and bromo substitutions on benzene ring did not exhibit much activity. Compound **4a** showed least activity, because it has no substituent on the benzene ring. However, the degree of inhibition varied both with the test compound, as well as with the bacteria used in the present investigation.

In conclusion, the antibacterial activity of compounds **4b & 4d** is promising compared to standard drug *Ciprofloxacin*, and they can be exploited for formulation of bactericides after further study.

Table 1. Antibacterial activity of 4-(1*H*-benzo[*d*]imidazol-2-yl)-8,9-diphenyl-1,6-dioxa-4,7,9- triazaspiro[4,5]dec-7-en-3-ones **4a-h**

Compound	Minimum Inhibitory Concentration in µg/ml (MIC) [a,b]					
	Gram + ve bacteria			Gram –ve bacteria		
	B.subtilis	*B.sphaericus*	*S.aureus*	*P.aeruginosa*	*K.aerogenes*	*C.violaceum*
4a	13	11	11	12	12	13
4b	9	8	9	7	10	8
4c	18	16	16	20	15	18
4d	10	9	8	8	9	10
4e	16	15	15	17	16	15
4f	8	9	6	8	7	9
18	18	18	20	22	18	20
4h	16	14	14	13	13	15
Ciprofloxacin	20	22	26	25	20	22

[a]Negative control (acetone) – No activity
[b]Values are indicated in µg/mL

Antifungal activity

The newly synthesized 4-(1*H*-benzo[*d*]imidazol-2-yl)-8,9-diphenyl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-ones (**4a-h)** were evaluated for their antifungal activity against *Fusarium oxysporum*, *Verticillium dahliae*, *Alternaria solani*, *Rhizoctonia solani*, *Colletotrichum capsici* and *Pythium aphanidermatum* in acetone by agar cup bioassay method[xvii], using *Flucanozole* as the standard drug.

Antifungal activity data (**Table 2**) revealed that compounds **4a-h** highly toxic towards all the fungi under investigation. Compounds **4b** and **4d** exhibited high antifungal activity by inhibiting the growth of fungi to a remarkable extent, when compared to standard drug *Fluconazole*, which may be due to the presence of methyl, methoxy substituents on the benzene ring, besides the presence of basic skeleton. Compound **4a** showed good activity. Compounds **4c, 4e and 4g** are moderately active. However, the degree of spore germination inhibition varied with the test compound as well as with the fungi under investigation. It is noteworthy that compounds **4b and 4d** showed better activity, when compared with the standard drug *Fluconazole*, hence, they may be exploited for control of wilt diseases of different crops as fungicides after further studies.

Table 2. Antifungal activity of 4-(1*H*-benzo[*d*]imidazol-2-yl)-8,9-diphenyl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-ones **4a-h**

Compd.N0.	Minimum inhibitory Concentration in µg/mL (MIC)[a,b]					
	F.oxysporum	*V. dahliae*	*A. solani*	*R. solani*	*C. capsici*	*P. aphanidermatum*
4a	15	15	18	16	16	20
4b	11	10	11	10	9	10
14	14	15	17	15	16	18
4d	9	11	10	10	11	9
4e	15	15	16	16	14	20
4f	8	9	9	10	11	11
4g	13	14	18	14	15	16
4h	15	13	18	15	16	19
Fluconazole	16	16	20	16	18	22

[a]Negative control (acetone) – No activity**.**
[b]Values are indicated in µg/mL

Experimental Section

All the melting points were determined on a Fisher-Johns melting point apparatus and are uncorrected. Analytical TLC was performed on Merck precoated 60 F_{254} silica gel plates. Visualization was carried out by exposure to iodine vapour. IR spectra (KBr pellet) were recorded on a Perkin-Elmer BX series FT-IR spectrometer. ^{1}H NMR spectra were recorded on a Bruker 300 MHz spectrometer. ^{13}C NMR spectra were recorded on a Bruker 75 MHz spectrometer. Chemical shift values are given in δ ppm with tetramethyl silane as an internal standard. ESI Mass spectra were recorded on a Agilent LC-MSD mass spectrometer. Elemental analyses were performed on a Carlo Erba 106 and Perkin-Elmer model 240 analyzers.

General procedure for the Synthesis of *N*-(1*H*-benzo[*d*]imidazol-2-yl)-2-chloroacetamide (2)

2-Aminobenzimidazole (**1)** (0.01 mol), chloroacetyl chloride (0.01 mol) and triethyl amine (0.5 mL) were taken in dry benzene. The contents were refluxed while stirring for 4 h. The precipitated triethylamine hydrochloride was removed by filtration. The gummy product obtained after the removal of solvent at ambient temperature was triturated with methanol. Recrystallization of the product was effected from aqueous alcohol. m.p. 130 ^{0}C. Yield 80%. Anal. Calcd. For $C_9H_8ClN_3O$. C; 51.67, H; 3.82, N; 20.09; Found: C; 51.64, H; 3.85, N; 20.04%

General procedure for the Synthesis of 3-(1*H*-benzo[*d*]imidazol-2-yl)-2-(phenylimino)oxazolidin-4-one (3a-h)

A mixture of *N*-(1*H*-benzo[*d*]imidazol-2-yl)-2-chloroacetamide **(2)** (0.01 mol), aryl isocyanate (0.01 mol), and K_2CO_3 (0.5 g) was taken in acetonitrile (15 mL). The reaction mixture was stirred at room temperature for about 6 h. The solvent was removed under reduced pressure and the residue was purified by recrystallization from ethanol.

3-(1*H*-Bnzo[*d*]imidazol-2-yl)-2-(phenylimino)oxazolidin-4-one (3a):

Pale yellow solid (80%), mp 135-37^{o}C. IR: (KBr) cm^{-1} 1635 (C=N), 1690 (CO), 3345 (benzimidazole-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δppm: 4.22 (s, 2H, CH_2), 6.98 -7.70 (m, 9H, Ar-H), 12.25 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm:72.54, 115.38, 117.74, 124.36, 125.64, 127.98, 129.47, 131.34, 133.37, 134.47, 142.72, 143.54, 150.28, 154.49, 179.42, 190.21; ESI-MS: *m/z* 293 $[M+H]^+$. Anal. Calcd. for $C_{16}H_{12}N_4O_2$: C, 65.75; H, 4.10; N, 19.17. Found: C, 65.78; H, 4.14; N, 19.14%.

2-(4-Methoxyphenylimino)-3-(1*H*-benzo[*d*]imidazol-2-yl)oxazolidin-4-one (3b):

Pale yellow solid (84%), mp 145-47^{o}C. IR: (KBr) cm^{-1} 1632 (C=N), 1688 (CO), 3340 (benzimidazole-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δppm: 3.60 (s, 3H, OCH_3), 4.25 (s, 2H, CH_2), 6.99 -7.79 (m, 8H, Ar-H), 12.29 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 46.37, 73.84, 118.78, 119.24, 126.58, 127.69, 129.98, 131.57, 133.44, 134.39, 135.47, 143.70, 144.54, 152.58, 156.29, 181.44, 192.47; ESI-MS: *m/z* 323 $[M+H]^+$. Anal. Calcd. for $C_{17}H_{14}N_4O_3$: C, 63.35; H, 4.34; N, 17.39. Found: C, 63.38; H, 4.36; N, 17.35%.

2-(4-Chlorophenylimino)-3-(1*H*-benzo[*d*]imidazol-2-yl)oxazolidin-4-one (3c):

Pale yellow solid (75%), mp 151-53^{o}C. IR: (KBr) cm^{-1} 1637 (C=N), 1692 (CO), 3347 (benzimidazole-NH); ^{1}H NMR (300 Hz, $CDCl_3$) δppm: 4.24 (s, 2H, CH_2), 6.99 -7.75 (m, 8H, Ar-H), 12.26 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 73.74, 116.38, 117.74, 125.29, 126.84, 128.65, 130.41, 132.22, 134.67, 135.55, 143.82, 144.60, 151.58, 155.43, 180.02, 191.11; ESI-MS: *m/z* 327 $[M+H]^+$. Anal. Calcd. for $C_{16}H_{11}N_4O_2Cl$: C, 58.89; H, 3.37; N, 17.17. Found: C, 58.86; H, 3.34; N, 17.14%.

2-(*P*-tolylimino)-3-(1*H*-benzo[*d*]imidazol-2-yl)oxazolidin-4-one (3d):

Pale yellow solid (83%), mp 139-41^{o}C. IR: (KBr) cm^{-1} 1631 (C=N), 1687 (CO), 3341 (benzimidazole-NH) cm^{-1}; ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 2.45 (s, 3H, CH_3), 4.24 (s, 2H, CH_2), 6.99 -7.80 (m, 8H, Ar-H), 12.28 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 23.45, 72.74, 117.88, 118.20, 125.48, 126.60, 128.98, 130.53, 132.70, 133.39, 134.77, 142.79, 143.34, 151.88, 155.09, 180.64, 191.22; ESI-MS: *m/z* 307 $[M+H]^+$. Anal. Calcd. for $C_{17}H_{14}N_4O_2$: C, 66.66; H, 4.57; N, 18.30. Found: C, 66.64; H, 4.55; N, 18.34%.

2-(4-Bromophenylimino)-3-(1*H*-benzo[*d*]imidazol-2-yl)oxazolidin-4-one (3e):

Brown solid (74%), mp 189-91^{o}C. IR: (KBr) cm^{-1} 1636 (C=N), 1691 (CO), 3345 (benzimidazole-NH) cm^{-1}; ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 4.22 (s, 2H, CH_2), 6.94 -7.73 (m, 8H, Ar-H), 12.25 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 72.54, 115.28, 116.74, 125.21, 125.84, 127.55, 130.48, 131.52, 133.69, 135.59, 142.72, 144.53, 150.58, 154.42, 180.06, 190.13; ESI-MS: *m/z* 371 $[M+H]^+$. Anal. Calcd. for $C_{16}H_{11}N_4O_2Br$: C, 51.89; H, 2.97; N, 15.13. Found: C, 51.86; H, 2.96; N, 15.10%

2-(4-Nitrophenylimino)-3-(1*H*-benzo[*d*]imidazol-2-yl)oxazolidin-4-one (3f):

Brown solid (74%), mp 180-82^{o}C. IR: (KBr) cm^{-1} 1639 (C=N), 1696 (CO), 3350 (benzimidazole-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 4.29 (s, 2H, CH_2), 6.99 -7.89 (m, 8H, Ar-H), 12.28 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 75.34, 118.18, 119.44, 128.25, 128.98, 130.55, 133.68, 134.32, 136.76, 138.59, 145.32,

147.63, 153.50, 157.52, 183.14, 193.43; ESI-MS: *m/z* 338 $[M+H]^+$. Anal. Calcd. for $C_{16}H_{11}N_5O_4$: C, 56.97; H, 3.26; N, 20.77. Found: C, 56.95; H, 3.24; N, 20.74%

2-(4-(Dimethylamino)phenylimino)-3-(1*H*-benzo[*d*]imidazol-2-yl)oxazolidin-4-one (3g):

Pale yellow solid (86%), mp 165-67^{o}C. IR: (KBr) cm^{-1} 1636 (C=N), 1689 (CO), 3341 (benzimidazole-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 2.54 (s, 6H, Ar-N$(CH_3)_2$), 4.25 (s, 2H, CH_2), 6.99 -7.90 (m, 8H, Ar-H), 12.29 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 24.45, 48.76, 49.87, 73.74, 118.98, 119.20, 127.50, 129.98, 131.33, 133.50, 134.39, 135.67, 143.79, 144.34, 152.48, 156.09, 181.64, 192.12; ESI-MS: *m/z* 336 $[M+H]^+$. Anal. Calcd. for $C_{18}H_{17}N_5O_2$: C, 64.47; H, 5.07; N, 20.89. Found: C, 64.44; H, 5.04; N, 20.85%.

2-(2,4-Dichlorophenylimino)-3-(1*H*-benzo[*d*]imidazol-2-yl)oxazolidin-4-one (3h):

Pale yellow solid (74%), mp 175-77^{o}C. IR: (KBr) cm^{-1} 1638 (C=N), 1693 (CO), 3348 (benzimidazole-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 4.26 (s, 2H, CH_2), 6.99 -7.83 (m, 7H, Ar-H), 12.28 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 74.74, 117.28, 118.74, 126.39, 127.88, 129.43, 131.61, 133.42, 135.67, 136.55, 144.89, 145.62, 152.50, 156.83, 181.00, 192.19; ESI-MS: *m/z* 361 $[M+H]^+$.Anal. Calcd. for $C_{16}H_{10}N_4O_2Cl_2$: C, 53.33; H, 2.77; N, 15.55. Found: C, 53.36; H, 2.74; N, 15.59%.

General procedure for the synthesis of 4-(1*H*-benzo[*d*]imidazol-2-yl)-8,9-diphenyl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-one (4a-h)

To a solution of 3-(1*H*-benzo[*d*]imidazol-2-yl)-2-(arylimino)-oxazolidin-4-ones **(3)** (0.01 mol) in dry chloroform (50 mL) in ice-salt bath, benzhydroxamoyl chloride (0.01 mol) in chloroform (10 mL) was added. Triethylamine (0.01 mol) in chloroform (20 mL) was added to the reaction mixture at 0 ^{o}C during 15 min with constant stirring. After the addition was complete the stirring was continued for another 4 h at 0^{0}C. The chloroform layer was washed with water (2 X 25 mL) to make it free form triethyl amine hydrochloride and the organic layer was dried (Na_2SO_4). The solvent was removed at ambient temperature and crude product was triturated with light petrol repeatedly to obtain a residue. The residue on shaking with methanol gave the solid which was recrystallized from benzene.

4-(1*H*-Benzo[*d*]imidazol-2-yl)-8,9-diphenyl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-one (4a):

Pale yellow solid (80%), mp 158-60^{o}C. IR: (KBr) cm^{-1} 1610 (C=N), 1655 (CO), 3456 (-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 4.10 (s, 2H, CH_2), 7.10 -7.83 (m, 14H, Ar-H), 12.25 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 52.56, 62.46, 114.54, 115.67, 116.62, 117.49, 118.48, 119.83, 123.83, 124.76, 127.86, 128.39, 129.73, 130.64, 132.87, 134.98, 135.39, 137.87, 138.28, 145.87, 150.38, 180.58, 190.54; ESI-MS: *m/z* 412 $[M+H]^+$. Anal. Calcd. for $C_{23}H_{17}N_5O_3$: C, 67.15; H, 4.13; N, 17.03. Found: C, 67.18; H, 4.15; N, 17.07%.

4-(1*H*-Benzo[*d*]imidazol-2-yl)-9-(4-methoxyphenyl)-8-phenyl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-one (4b):

Pale yellow solid (85%), mp 173-75^{o}C. IR: (KBr) cm^{-1} 1614 (C=N), 1657 (CO), 3460 (-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 3.68 (s, 3H, Ar-OCH_3), 4.12 (s, 2H, CH_2), 7.11 -7.85 (m, 13H, Ar-H), 12.28 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 48.87, 54.76, 64.66, 116.58, 117.57, 118.67, 119.50, 120.78, 121.83, 125.95, 126.76, 129.26, 130.79, 131.73, 132.64, 134.81, 136.99, 137.30, 139.67, 140.28, 147.37, 152.38, 182.88, 192.24; ESI-MS: *m/z* 412$[M+H]^+$. Anal. Calcd. for $C_{24}H_{19}N_5O_4$: C, 65.30; H, 4.30; N, 15.87. Found: C, 65.33; H, 4.27; N, 15.85%.

4-(1*H*-Benzo[*d*]imidazol-2-yl)-9-(4-chlorophenyl)-8-phenyl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-one (4c):

Pale yellow solid (75%), mp 189-91^{o}C. IR: (KBr) cm^{-1} 1613 (C=N), 1654 (CO), 3458 (-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 4.12 (s, 2H, CH_2), 7.10 -7.85 (m, 13H, Ar-H), 12.26 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 53.16, 63.66, 115.54, 116.87, 117.62, 118.40, 119.48, 120.89, 124.33, 125.86, 128.80, 129.39, 130.83, 131.69, 133.37, 135.98, 136.39, 138.57, 139.20, 146.83, 151.39, 181.93, 191.24; ESI-MS: *m/z* 446 $[M+H]^+$. Anal. Calcd. for $C_{23}H_{16}N_5O_3Cl$: C, 62.09; H, 3.59; N, 15.73. Found: C, 62.07; H, 3.57; N, 15.77%.

4-(1*H*-Benzo[*d*]imidazol-2-yl)-8-phenyl-9-(p-tolyl)-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-one. (**4d**):

Pale yellow solid (84%), mp 168-70^{o}C. IR: (KBr) cm^{-1} 1613 (C=N), 1656 (CO), 3455 (-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 2.45 (s, 3H, Ar-CH_3), 4.11 (s, 2H, CH_2), 7.10 -7.84 (m, 13H, Ar-H), 12.27 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 28.47, 53.66, 63.69, 115.48, 116.77, 117.67, 118.60, 119.78, 120.83, 124.55, 125.76, 128.26, 129.79, 130.73, 131.44, 133.81, 135.90, 136.30, 138.77, 139.08, 146.37, 151.33, 181.81, 191.24; ESI-MS: *m/z* 426 $[M+H]^+$. Anal. Calcd. for $C_{24}H_{19}N_5O_3$: C, 67.76; H, 4.47; N, 16.47. Found: C, 67.73; H, 4.44; N, 16.45%.

4-(1*H*-Benzo[*d*]imidazol-2-yl)-8-(4-chlorophenyl)-9-phenyl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-one (4e):

Pale yellow solid (74%), mp 184-86^{o}C. IR: (KBr) cm^{-1} 1612 (C=N), 1655 (CO), 3460 (-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 4.11 (s, 2H, CH_2), 7.10 -7.84 (m, 13H, Ar-H), 12.27 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 54.19, 64.69, 115.64, 117.80, 118.62, 119.40, 119.86, 121.89, 125.93, 126.16, 128.89, 129.39, 131.89, 132.60, 134.37, 136.98, 137.30, 138.57, 139.28, 147.83, 152.39, 180.99, 192.20; ESI-MS: *m/z* 446 $[M+H]^+$. Anal. Calcd. for $C_{23}H_{16}N_5O_3Cl$: C, 62.09; H, 3.59; N, 15.73. Found: C, 62.05; H, 3.55; N, 15.70%.

4-(1*H*-Benzo[*d*]imidazol-2-yl)-8-(4-methoxyphenyl)-9-phenyl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-one (4f):

Pale yellow solid (83%), mp 181-83^{o}C. IR: (KBr) cm^{-1} 1615 (C=N), 1659 (CO), 3462 (-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 3.69 (s, 3H, Ar-OCH_3), 4.11 (s, 2H, CH_2), 7.10 -7.89 (m, 13H, Ar-H), 12.28 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 49.87, 55.46, 65.60, 115.58, 118.77, 119.37, 119.99, 121.38, 122.89, 126.35, 127.70, 129.85, 131.72, 133.73, 134.64, 135.89, 136.93, 138.33, 139.69, 141.20, 148.31, 153.30, 182.93, 193.04; ESI-MS: *m/z* 442$[M+H]^+$. Anal. Calcd. for $C_{24}H_{19}N_5O_4$: C, 65.30; H, 4.30; N, 15.87. Found: C, 65.28; H, 4.33; N, 15.89%.

4-(1*H*-Benzo[*d*]imidazol-2-yl)-9-(4-bromophenyl)-8-phenyl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-one (4g):

Brown solid (72%), mp 202-02^{o}C. IR: (KBr) cm^{-1} 1613 (C=N), 1657 (CO), 3457 (-NH) ; ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 4.10 (s, 2H, CH_2), 7.10 -7.88 (m, 13H, Ar-H), 12.26 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 53.69, 65.61, 114.34, 116.80, 119.69, 118.48, 118.80, 121.89, 125.90, 126.54, 127.49, 128.79, 130.89, 131.69, 133.39, 135.98, 1376.39, 137.54, 138.58, 146.82, 151.30, 181.99, 191.21; ESI-MS: *m/z* 490 $[M+H]^+$. Anal. Calcd. for $C_{23}H_{16}N_5O_3Br$: C, 56.44; H, 3.27; N, 14.31. Found: C, 56.40; H, 3.30; N, 14.35%.

4-(1*H*-Benzo[*d*]imidazol-2-yl)-8-(4-nitrophenyl)-9-phenyl-1,6-dioxa-4,7,9-triazaspiro[4,5]dec-7-en-3-one (4h):

Pale yellow solid (74%), mp 194-96^{o}C. IR: (KBr) cm^{-1} 1616 (C=N), 1660 (CO), 3463 (-NH); ^{1}H NMR(300 Hz, $CDCl_3$) δ ppm: 4.14 (s, 2H, CH_2), 7.10 -7.90 (m, 13H, Ar-H), 12.28 (s, 1H, benzimidazole-H, D_2O exchangeable); ^{13}C NMR (75 MHz, $CDCl_3$) δ ppm: 53.86, 63.76, 115.59, 116.69, 117.82, 118.49, 119.48, 120.83, 124.30, 125.46, 128.86, 129.30, 130.77, 131.64, 133.83, 135.98, 136.30, 138.82, 139.28, 146.80, 151.38, 181.50, 191.58; ESI-MS: *m/z* 457 $[M+H]^+$. Anal. Calcd. for $C_{23}H_{16}N_6O_5$: C, 60.52; H, 3.50; N, 18.42. Found: C, 60.55 H, 3.57; N, 18.45%.

Acknowledgements

The authors are thankful to the Head, Department of Chemistry, Kakatiya University, Warangal for facilities and to the Director, CSIR- Indian Institute of Chemical Technology, Hyderabad for recording ^{1}H NMR, ^{13}C NMR and Mass Spectra.

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Received on April 14, 2018.