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Synthesis and antihypertensive activity of Schiff bases of 4'-(6-chloro-5-nitro-2-[4-(3-substituted-phenyl-acryloylamino)-phenyl]-benzimidazole-1-yl-methyl)-biphenyl-2-carboxylic acids

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ABSTRACT

New Series of Antihypertensive agents Schiff bases 4'-(6-chloro-5-nitro-2-[4-(3-substituted-phenyl-acryloylamino)-phenyl]-benzimidazole-1-ylmethyl)-biphenyl-2-carboxylic acid and Side chain in the using different aromatic aldehydes have been synthesized from substituted compounds [1-10] and tested for antihypertensive activity in induced hypertensive rats. All the compounds have been found to be less active than Losartan; their structures were assigned with elemental analysis, melting point and spectral analysis like IR, ^{1}H NMR, ^{13}C NMR and FAB Mass.

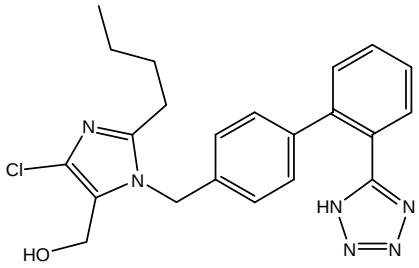
Keywords: angiotensin II, antihypertensive agents, Schiff bases, biphenyl-2-carboxylic acid.

INTRODUCTION

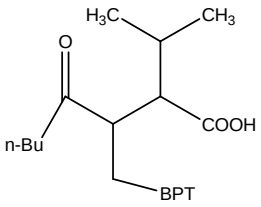
Hypertension is a common problem facing man today. Because high blood pressure is one of the leading causes of stroke and a major risk for heart attack, one of the most important aspects of preventive cardiology should be to identify who has the disease in many people as possible and to take steps to lower the blood pressure before it causes damage to the blood vessels, heart, kidney, eyes and other organs [1, 2] So, hypertension (high blood pressure), is a condition commonly associated with narrowing of the arteries. This causes blood to be pumped with excessive force against the artery walls. It is called “Silent killer” because most people have no reason to think they might be hypertensive [3, 4].The renin-angiotensin system (RAS) plays an important role in blood pressure control and in water and salt homeostasis which control the pathophysiology of a number of cardiovascular disorders such as malignant hypertension [5]. Originally, RAS was regarded as a circulating hormone system. Recent studies, however, have demonstrated the existence of local RAS in many tissues including the brain, kidney, adrenal cortex, heart and blood vessel wall [6]. Tissue RAS is activated in pathophysiological situations and local synthesis of Ang II appears to contribute to altered tissue function and morphology [7].The renin-angiotensin system (RAS) plays an important role in the regulation of blood

pressure through the actions of angiotensin II (AII) (vasoconstriction, aldosterone secretion, renal sodium re-absorption, and nor epinephrine release) and thus is an appropriate target for therapeutic intervention in hypertension. The renin-angiotensin system (RAS) plays a key role in regulating cardiovascular homeostasis and electrolyte/ fluid balance in normotensive and hypertensive subjects.[8]Activation of the renin-angiotensin cascade begins with rennin secretion from the juxtaglomerular apparatus of the kidney and culminates in the formation of the octapeptide angiotensin II (AII), which then interacts with specific receptors present in different tissues.[9] Two basic types of receptors, both having a broad distribution, have been characterized so far: the AT1 receptor, responsible for the majority of effects attributed to this peptide, and the AT2 receptor, with a functional role yet uncertain.[10] The main effects of AII are the regulation of blood pressure through vasoconstriction, thereby effecting an increase in vascular resistance, the regulation of volemia through the stimulated release of vasopressin and aldosterone, which induces saline retention, and the regulation of the adrenocorticotropic hormone (ACTH). Thus, reducing the levels of AII by inhibition of one of the RAS enzymes or directly blocking the AII receptors is in theory a good approach for treating hypertension, confirmed by the success of angiotensin-converting enzyme (ACE) inhibitors as antihypertensive [11].Substantial effort has been made to find renin inhibitors, although orally active agents have only recently been reported[12].The discovery of potent and orally active nonpeptide Ang II antagonists such as losartan and eprosartan has encouraged the development of a large number of similar compounds[13]. Among them, irbesartan, candesartan, valsartan, telmisartan, tasosartan,and olmesartan are on the market. Most of the developed AT_1 receptor antagonists are characterized by the presence in their structure of the biphenyl fragment bearing an acidic moiety and differ in the nature of the pendent heterocyclic system (valsartan lacks the heterocyclic moiety) connected to the Para position of the proximal phenyl.Substantial effort has been made to find renin inhibitors, although orally active agents have only recently been reported [14]. No less effort has been devoted to finding AII antagonists, which besides being the most direct way of controlling the RAS could have the additional advantage of lacking the side effects, such as cough and angioedema, observed with ACE inhibitors, as these are probably caused by partial inhibition of the cleavage of bradykinin and substance P. Starting from the initial leads reported by Takeda [15]. Researchers at DuPont discovered losartan, the first orally active AT1 selective nonpeptide AII antagonist that reached the market for the treatment of hypertension (1994, Cozaar). Whereas reports on effective replacements of the biphenyl tetrazole “tail” of losratan are scarce, the imidazolic “head” of the molecule, postulated to act mainly to link the required functionalities, has been successfully replaced by a wide variety of cyclic and acyclic structures, leading to a number of compounds currently in clinical trials.[16].AngII receptor antagonists are expected to have similar therapeutic effects and indications as the ACE inhibitors without unwanted side effects associated inhibition of other ACE mediated pathways, such as bradykinin metabolism.Intial research in this area led to the discovery of peptide analog such as saralasin ([sar1-Ala8]-AngII) which displayed potent and selective AngII receptor antagonist activity both *in vivo and in vitro*.However, these peptides had limited therapeutic utility due to partial agonist activity short duration of action and lack of appreciable oral bioavailability[17].Only in recent years a number of non peptides AngII antagonists that show promise as inhibitors of the RAS been reported[18].All these antagonists possess a central aromatic nucleus bearing the pharmacophores indispensable for activity and notably a polar function adjustant to biphenyl substituents while a polar function in this area of molecule seems to be necessary to maintain activity[19]. Sartans are appropriately substituted heterocyclic head coupled through a methylene linker to pendent biphenyl system bearing an acidic function; viz. candesartan is an effective competitive Ang II antagonist with benzimidazole nucleus as the heterocyclic head [20]. The substitute at 6-position on the nucleus increases the activity whereas small substituent at 5-position decreases the activity [21]. Compounds containing tetrazole nucleus are also reported as

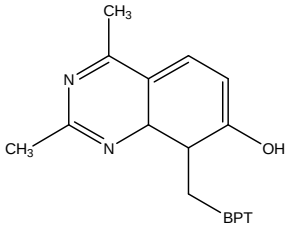
AT_1 receptor antagonists and their protypical derivative 3 exhibits non-competitive antagonism[22] and amino group attach with carboxylic group given good biological activity [23-25].



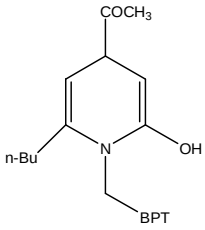
Losartan



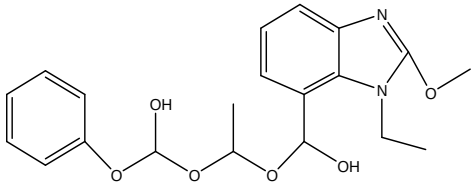
Valsartan



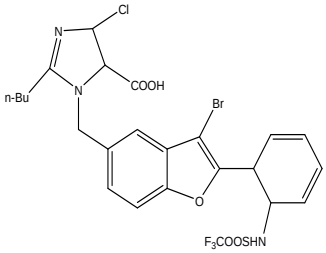
Tasosartan



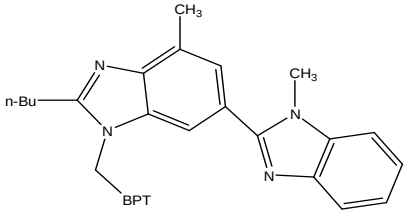
Candesartan



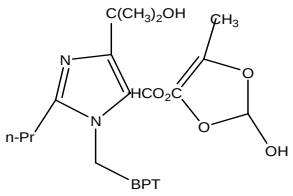
Milfasartan



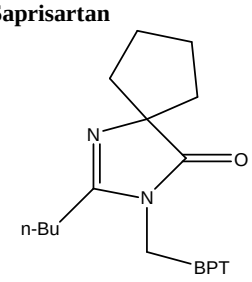
Saprisartan



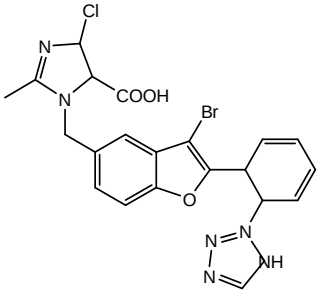
Telmisartan



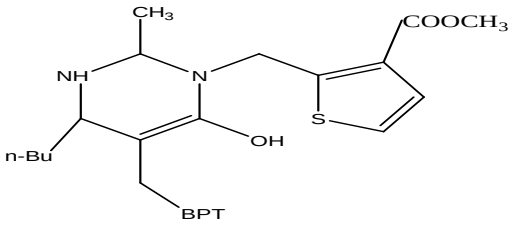
Olmesartan



Irbesartan



Zolzsartan



Eprosartan

Angiotensin II selective antagonists family

MATERIALS AND METHODS

Melting points were determined in open capillary tubes and are uncorrected. The time required for completion of the reaction was monitored by TLC using Silica gel-G plates and spots were exposed in iodine chamber. IR spectra were recorded on a Perkin Elmer 1800 (FTIR) spectrometer 1H NMR spectra (DMSO) were taken on a DRX-300 spectrometer (300 MHz) using TMS as internal standard and chemical shifts are expressed in δ ppm.

***MS-01-Synthesis of* 4-(6-Chloro-1H-benzoimidazol-2-yl)-phenyl amine**

A solution of 4-Chloro-1,2-phenylenediamine dihydrochloride (0.45 g, 2.5 mmol) in 5 ml of

water was cooled to 0°C and treated with a solution of cyanogen bromide (0.60 ml, 5 M in acetonitrile, 3.0 mmol) and solid $NaHCO_3$ (0.41 mg, 4.9 mmol). The solution was stirred at ambient temperature for 40-45 h. The mixture was made basic with 1 M aqueous Na_2CO_3 and the solution was concentrated under reduced pressure. The residue was triturated with hot ethanol, and the ethanolic solution was filtered and concentrated under reduced pressure to obtain the compound **1** in appreciable yield.

Yield 80%; mp 142-145 ^{0}C; Anal Calcd for $C_{13}H_{10}ClN_3$ (R=H): C, 64.04; H, 4.14; N, 17.24%; IR (υ cm^{-1}): 3045 (C-H, sp^2), 3210 (NH, bonded), 3175 (NH, free), 1654 (C=N), 1626, 1586, 1444 (C⋯C, ring str) 958, 859, 742 (sub. phenyl) 647 (C-Cl); ^{1}H NMR (300 MHz, $CDCl_3$) δ: 4.0 (s, 2H, NH_2), 5.0 (s,1H, NH), 7.3-7.8 (m, 7H, Ar-H); ^{13}C NMR ($CDCl_3$) δ: 111.6,115.1,119.7,126.1,141.6; FAB-MS: 243.692 $(M+H)^+$.

MS-02-4-(6-Chloro -5-nitro-1H-benzoimidazol-2-yl)-phenyl amine

Twenty ml of concentrated nitric acid was placed in three necked flask and equal quantity of concentrated sulphuric acid (1:1) was added slowly. The mixture was kept in the ice cold water then compound (different R-aryl groups) (15.10 gm) was mixed in portions during 2 hour under room temperature. After stirred continuously for 8 hrs hours minutes and then the reaction mixture was poured slowly over crushed ice with stirring. The precipitated product was filtered out and washes with cold water. The final product recrystillzed from absolute ethanol.

Yield 82%; mp 135-138 ^{0}C; Anal Calcd for $C_{13}H_9N_4O_2$ (R=H): C, 54.09; H, 3.17; N, 19.41%; IR (υ cm^{-1}): 3041 (C-H, sp^2), 3216 (NH, bonded), 3171 (NH, free), 1653 (C=N), 1654 (NO_2),1629, 1589, 1449 (C⋯C, ring str) 952, 866, 773 (sub. phenyl), 649 (C-Cl); ^{1}H NMR (300 MHz, $CDCl_3$) δ: 4.05 (s, 2H, NH_2), 5.01 (s, NH), 7.4-7.9 (m, 6H, Ar-H); ^{13}C NMR ($CDCl_3$) δ: 110.1,113.5,118.6,121.3, 131.2, 142.2; FAB-MS: 288.04 $(M+H)^+$.

MS-03-N-[4-(6-Chloro -5-nitro-1H-benzoimidazol-2-yl)-phenyl]acetamide

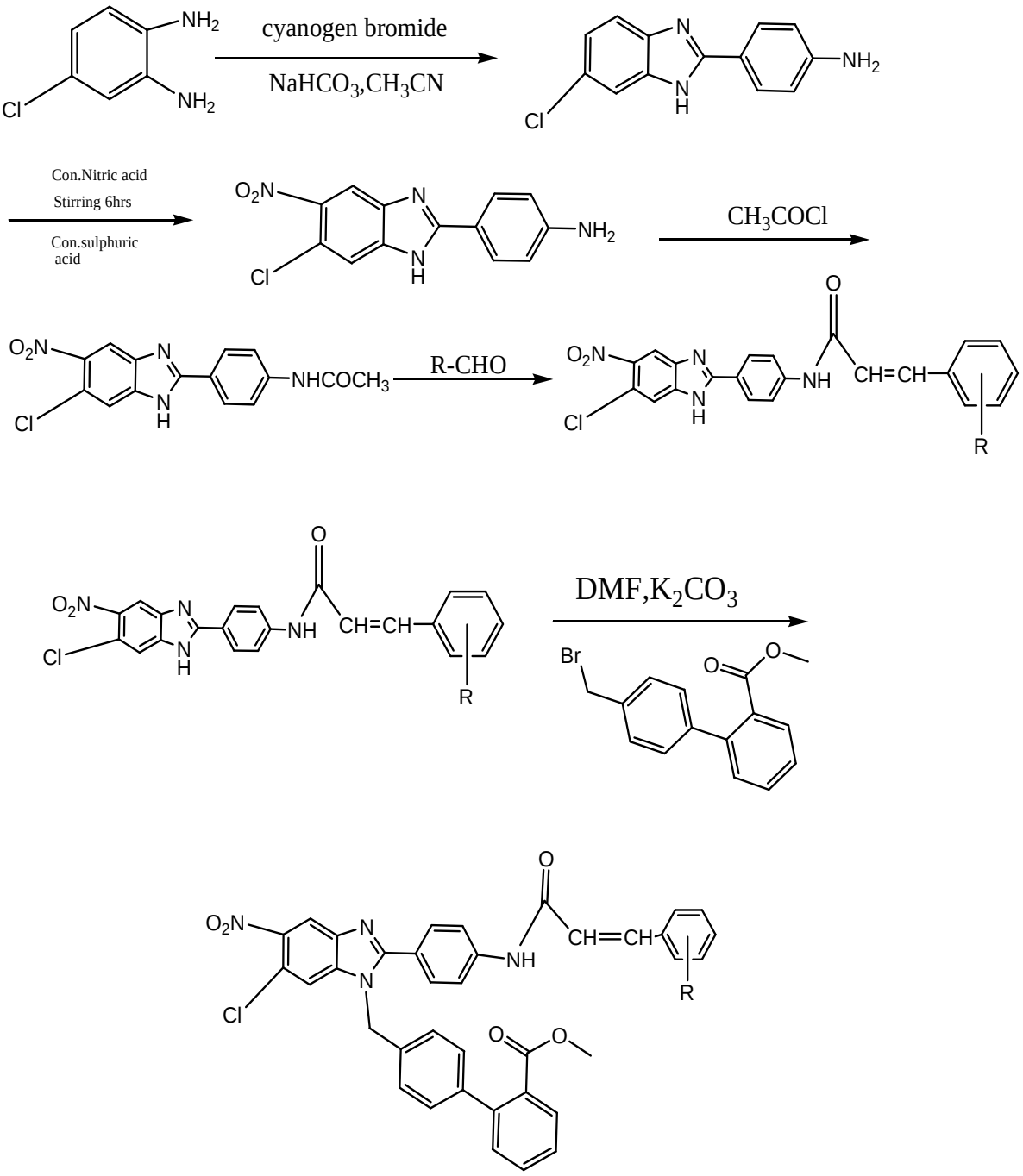
Dissolve 4-(6-Chloro -5-nitro-1H-benzoimidazol-2-yl)-phenyl amine (1.5 g, 0.01 mole) in absolute ethanol (100 mL) and acetyl chloride (1.5 g, 0.01 mole) was added drop wise with constant stirring at 0-5^{0}C. The reaction mixture was stirred for 6 hrs. The excess solvent was distilled off and the solid product was filtered, dried and recrystallised from ethanol to give compound yield

Yield 70 %; mp 156-158 ^{0}C; Anal Calcd for $C_{15}H_{11}N_4O_3$ (R=H): C, 54.49; H, 3.37; N, 16.94%; IR (υ cm^{-1}): 3045 (C-H, sp^2), 3210 (NH, bonded), 3179 (NH, free), 1652 (C=N), 1651(NO_2),1625, 1580, 1442 (C⋯C, ring str) 956, 861, 770 (sub. phenyl), 646 (C-Cl); ^{1}H NMR (300 MHz, $CDCl_3$) δ: 4.01 (s, 2H, NH_2), 5.0 (s, NH), 7.2-7.7 (m, 6H, Ar-H),2.02(s3H,methyl; ^{13}C NMR ($CDCl_3$) δ: 111.2,112.1,115.8,127.5,139.8; FAB-MS: 330.05 $(M+H)^+$.

MS-04-N-[4-(6-Chloro -5-nitro-1H-benzoimidazol-2-yl)-phenyl]acetamide

Dissolve N-[4-(6-Chloro -5-nitro-1H-benzoimidazol-2-yl)-phenyl]acetamide (1.12g, 0.01 mole) in absolute ethanol (30 mL) and various aromatic aldehydes (1.06 g, 0.01 mole) were taken and then an aqueous solution of KOH (2%, 5 mL) added to it. The reaction mixture refluxed for 5 h and then the solvent was removed by vacuum distillation and then it was poured into crushed ice and acidified with HCl. The solid separated was filtered and recrystallised from ethanol (Scheme). Similarly remaining compounds **[1-10]** was prepared by above method.

SCHEME



Compound [1-10]

MS-05-Synthesis of 4'-(6-chloro-5-nitro-2-[4-(3-substituted-phenyl-acryloylamino)-phenyl]-benzimidazole-1-ylmethyl)-biphenyl-2-carboxylic acid

1.15gm of MS-04 was dissolved in 20ml of DMF (dimethyl formamide) and stirred vigorously with 1.5gm of potassium carbonate at 27^{0}C for four hour. To the resulting mixture 0.482gm of 4'bromomethylbiphenyl-2-carboxylic acid dissolved in 20 ml of DMF and then was added drop wise with dropping funnel in three hour the reaction was allowed to proceed for further 9 hours at room temperature and solvent removed under vacuum. Residue was treated with 20ml of

dilute HCl and extracted with ethyl acetate. The organic layer was washed with brine solution, distilled water and dried over anhydrous sodium sulphate. (MS-06) was obtained.

Spectral Data

[1]4'-(6-chloro-5-nitro-2-[4-(2-chloro-phenyl-acryloylamino)-phenyl]-benzimidazole-1-yl-methyl)-biphenyl-2-carboxylic acid

Yield: 75%, m.p. = 177^0-179^0C. Anal.Calcd for $C_{36}H_{24}Cl_2N_4O_5$: Found: C,65.17; H, 3.65; N, 8.44 %; IR (KBr): 3391(Broad O-H str.), 3055 (C-H, sp^2), 3219 (NH, bonded), 3167 (NH, free), 2810 (C-H str., CH_2), 1698 (carboxylic, C=O str.), 1607, 1640(CH=CH), 1541(C=N and C=Cstr.), 1530-1318 (N-O str., NO_2), 1140 (C-N str.), 654.4(C-Cl str); 1HNMR (300 MHz, $CDCl_3$); 10.54(s, 1H, COOH), 12.16(1H, s, NH-Benzimidazole); 7.1- 8.5 (m, 20H, ArH), 5.0(s, 2H, CH_2); ^{13}CNMR($CDCl_3$)δ: 58.3, 111.3, 112.1, 116.2, 127.1, 131.4, 133.1, 139.1, 142.2, 147.2, 152.5, FAB-MS, 662.12.

[2]4'-(6-chloro-5-nitro-2-[4-(2-bromo-phenyl-acryloylamino)-phenyl]-benzimidazole-1-yl-methyl)-biphenyl-2-carboxylic acid

Yield: 61%, m.p. = 243^0-245^0C. Anal.Calcd for $C_{36}H_{24}BrClN_4O_5$: Found: C, 61.08; H, 3.43; N, 7.92 %; IR (KBr): 3386(Broad O-H str.), 3068 (C-H, sp^2), 3225 (NH, bonded), 3163 (NH, free), 2818 (C-H str., CH_2),1723 (carboxylic, C=O str.), 1627, 1646(CH=CH), 1532(C=N and C=Cstr.), 1535-1311(N-O str., NO_2), 1154 (C-N str.), 651(C-Cl str); 1HNMR (300 MHz, $CDCl_3$), 10.59(s, 1H, COOH), 12.11(1H, s, NH-Benzimidazole), 7.04-8.64(m, 20H, ArH), 5.0(s, 2H, CH_2); ^{13}CNMR($CDCl_3$)δ: 50.1, 53.3, 65.1, 73.7, 112.4, 114.1, 116.1, 117.1, 122.2, 130.2, 141.1, FAB-MS, 706.46.

[3] 4'-(6-chloro-5-nitro-2-[4-(2-hydroxy-phenyl-acryloylamino)-phenyl]-benzimidazole-1-yl-methyl)-biphenyl-2-carboxylic acid

Yield: 65%, m.p.=222^0-225^0C. Anal. Calcd for $C_{36}H_{25}ClN_4O_6$: Found: C, 67.03; H, 3.91; N, 8.69%; IR (KBr): 3393(Broad O-H str.), 3073 (C-H, sp^2), 3229 (NH, bonded), 3168 (NH, free), 2812 (C-H str., CH_2), 1720 (carboxylic, C=O str.), 1620, 1665(CH=CH), 1538(C=N and C=Cstr.), 1530-1359 (N-O str., NO_2), 1146 (C-N str.), 650(C-Cl str); 1HNMR (300 MHz, $CDCl_3$), 10.43(s, 1H, COOH), 12.04(1H, s, NH-Benzimidazole), 7.1-8.33(m, 20H, ArH), 5.14(s, 1H, arm-OH), 4.97(s, 2H, CH_2); ^{13}CNMR($CDCl_3$)δ: 46.4, 53.2, 55.3, 67.1, 71.3, 112.2, 113.5, 117.1, 118.1, 124.1, 132.2, 134.1, 143.5; FAB-MS, 644.14.

[4]4'-(6-chloro-5-nitro-2-[4-(2-fluoro-phenyl-acryloylamino)-phenyl]-benzimidazole-1-ylmethyl)-biphenyl-2-carboxylic acid

Yield: 55%, m.p. = 264^0-266^0C. Anal.Calcd for $C_{36}H_{24}FClN_4O_5$: Found: C,66.82;H, 3.73;N,8.66 %;IR (KBr): 3375(Broad O-H str.), 3060(C-H, sp^2), 3236 (NH, bonded), 3176(NH, free), 2841 (C-H str., CH_2), 2818 (C-H str., CH_2),1716 (carboxylic, C=O str.),1614, 1631(CH=CH),1525(C=N and C=Cstr.), 1522-1362(N-O str., NO2), 1154 (C-N str.), 646(C-Cl str).1^H NMR (300 MHz, $CDCl_3$), 10.73(s, 1H, COOH), 12.02(1H, s, NH-Benzimidazole), 7.13-8.41 (m, 20H, ArH), 5.08(s, 2H, CH_2). ^{13}CNMR($CDCl_3$)δ: 48.0, 51.2, 53.3, 62.1, 73.8, 112.8, 113.4, 116.2, 117.2, 126.5, 130.1, 132.1, 140.6, FAB-MS, 647.16

[5]4'-(6-chloro-5-nitro-2-[4-(2-iodo-phenyl-acryloylamino)-phenyl]-benzimidazole-1-ylmethyl)-biphenyl-2-carboxylic acid

Yield: 67%, m.p. = 284^0-286^0C. Anal.Calcd for $C_{36}H_{24}IClN_4O_5$: Found: C,57,27;H, 3.21;N,7.43 %;IR (KBr): 3371(Broad O-H str.), 3052(C-H, sp^2), 3219 (NH, bonded), 3143(NH, free), 2839 (C-H str., CH_2), 2824(C-H str., CH_2),1711 (carboxylic, C=O str.), 1614, 1631(CH=CH), 1525(C=N and C=Cstr.), 1522-1362(N-O str., NO2), 1149 (C-N str.), 658(C-Cl str).1^H NMR

(300 MHz, $CDCl_3$), 10.87(s, 1H, COOH), 12.34(1H, s, NH-Benzimidazole), 7.2-8.5 (m, 20H, ArH), 5.11(s, 2H, CH_2). ^{13}CNMR($CDCl_3$)δ: 42.2, 112.3, 113.5, 117.2, 119.2, 125.5, 127.1, 136.1, 139.5, 144.5, FAB-MS, 754.04

[6]4'-(6-chloro-5-nitro-2-[4-(2-methoxy-phenyl-acryloylamino)-phenyl]-benzimidazole-1-ylmethyl)-biphenyl-2-carboxylic acid

Yield: 60%, m.p. = 205^0-207^{0}C. Anal.Calcd for $C_{37}H_{27}ClN_4O_6$: Found: C,67,43;H, 4.13;N,8.50 %;IR (KBr): 3356(Broad O-H str.), 3049(C-H, sp^2), 3242 (NH, bonded), 3158(NH, free), 2843 (C-H str., CH_2), 2814(C-H str., CH_2), 1716(carboxylic, C=O str.), 1619, 1622(CH=CH), 1544(C=N and C=Cstr.), 1546-1321(N-O str., NO_2), 1133 (C-N str.), 649(C-Cl str). 1^H NMR (300 MHz, $CDCl_3$), 10.76(s, 1H, COOH), 12.17(1H, s, NH-Benzimidazole), 7.2-8.5 (m, 20H, ArH), 5.06(s, 3H, OCH_3). 5.11(s, 2H, CH_2). ^{13}CNMR($CDCl_3$)δ: 42.2, 112.3, 113.5, 117.2, 119.2, 125.5, 127.1, 136.1, 139.5, 144.5, FAB-MS, 658.16

[7] 4'-(6-chloro-5-nitro-2-[4-(3-o-tolyl-phenyl-acryloylamino)-phenyl]-benzimidazole-1-ylmethyl)-biphenyl-2-carboxylic acid

Yield: 66%, m.p. = 185^0-187^{0}C. Anal.Calcd for $C_{37}H_{27}ClN_4O_5$: Found: C,69,10;H, 4.23;N,8.71%;IR (KBr): 3363(Broad O-H str.), 3051(C-H, sp^2), 3243 (NH, bonded), 3155(NH, free), 2968(t,3H,CH_3),2843 (C-H str., CH_2), 2814(C-H str., CH_2),1707 (carboxylic, C=O str.),1615, 1628(CH=CH),1540(C=N and C=Cstr.), 1546-1321(N-O str., NO_2), 1133 (C-N str.), 647.0(C-Cl str).1^H NMR (300 MHz, $CDCl_3$) 10.68(s,1H,COOH), 12.25(1H,s,-NH-Benzimidazole) 7.11- 8.5 (m,20H,ArH), 2.35(s,3H, CH_3). 5.14(s,2H,CH_2).^{13}CNMR($CDCl_3$)δ: 58.2,111.0,115.1,119.2,121.1,124.1,133.4,134.0,139.5,FAB-MS, 642.17

[8]4'-(6-chloro-5-nitro-2-[4-(3-chloro-phenyl-acryloylamino)-phenyl]-benzimidazole-1-ylmethyl)-biphenyl-2-carboxylic acid

Yield: 80%, m.p. = 171^0-173^{0}C. Anal.Calcd for $C_{36}H_{24}Cl_2N_4O_5$: Found: C,65.17;H, 3.65;N,8.44 %;IR (KBr): 3391(Broad O-H str.), 3055 (C-H, sp^2), 3219 (NH, bonded), 3167 (NH, free), 2810 (C-H str., CH_2),1698 (carboxylic, C=O str.),1607, 1640(CH=CH),1541(C=N and C=Cstr.), 1530-1318 (N-O str., NO2), 1140 (C-N str.), 654.4(C-Cl str).1^H NMR (300 MHz, $CDCl_3$) 10.54(s, 1H, COOH), 12.16(1H, s, NH-Benzimidazole), 7.1-8.5 (m, 20H, ArH), 5.0(s, 2H, CH_2).^{13}CNMR($CDCl_3$)δ: 58.3, 111.3, 112.1, 116.2, 127.1, 131.4, 133.1, 139.1, 142.2, 147.2, 152.5, FAB-MS, 662.12

[9]4'-(6-chloro-5-nitro-2-[4-(3-bromo-phenyl-acryloylamino)-phenyl]-benzimidazole-1-ylmethyl)-biphenyl-2-carboxylic acid

Yield: 66%, m.p. = 247^0-249^{0}C. Anal.Calcd for $C_{36}H_{24}BrClN_4O_5$: Found: C,61.08;H, 3.43;N,7.92 %;IR (KBr): 3386(Broad O-H str.), 3068 (C-H, sp^2), 3225 (NH, bonded), 3163 (NH, free), 2818 (C-H str., CH_2),1723 (carboxylic, C=O str.),1627, 1646(CH=CH),1532(C=N and C=Cstr.), 1535-1311 (N-O str., NO2), 1154 (C-N str.), 651(C-Cl str).1^H NMR (300 MHz, $CDCl_3$) 10.59(s, 1H, COOH), 12.11(1H, s, NH-Benzimidazole), 7.04-8.64 (m, 20H, ArH), 5.0(s, 2H, CH_2). ^{13}CNMR($CDCl_3$)δ: 50.1, 53.3, 65.1, 73.7, 112.4, 114.1, 116.1, 117.1, 122.2, 130.2, 141.1, FAB-MS, 707.75

[10]4'-(6-chloro-5-nitro-2-[4-(3-hydroxy-phenyl-acryloylamino)-phenyl]-benzimidazole-1-ylmethyl)-biphenyl-2-carboxylic acid

Yield: 60%, m.p.=226^0-229^{0}C. Anal. Calcd for $C_{36}H_{25}ClN_4O_6$: Found: C, 67.03; H, 3.91; N, 8.69%; IR (KBr): 3393(Broad O-H str.), 3073 (C-H, sp^2), 3229 (NH, bonded), 3168 (NH, free), 2812 (C-H str., CH_2), 1720 (carboxylic, C=O str.), 1620, 1665(CH=CH), 1538(C=N and C=Cstr.), 1530-1359 (N-O str., NO_2), 1146 (C-N str.), 650(C-Cl str). 1H NMR (300 MHz,

$CDCl_3$), 10.43(s, 1H, COOH), 12.04(1H, s, NH-Benzimidazole), 7.1-8.33 (m, 20H, ArH), 5.14(s, 1H, arm-OH), 4.97(s, 2H, CH_2); ^{13}CNMR($CDCl_3$)δ: 46.4, 53.2, 55.3, 67.1, 71.3, 112.2, 113.5, 117.1, 118.1, 124.1, 132.2, 134.1, 143.5; FAB-MS, 645.43.

Biological Activity[26-31]

Non-invasive Method (Indirect Method) Albino rats weighing 150-200 gm were used to screening for all the synthesizes benzimidazoles derivatives for antihypertensive activity. Suspension of test compound was prepared in 1% w/v sodium carboxy methyl cellulose and administered at dose level of 50 mg/kg animal body weight to different of five rats each group.Contorl group received an equal quantity of 1% w/v sodium carboxy methyl cellulose suspension. After administration of dose to animal, blood pressure was measured by Non-invasive Tail cuff Method using pressure meter.Measurment were done after 1 hour and 3 hour time interval intensive stepwise. One hour after administration of drug sample, animal was shifted to the restrainers, which restricts the movement of animal. The tail was cleaned with moist cotton to remove the dirty matter and talcum powder was sprayed on tail to make its surface smooth. A tail cuff and pulse transducer was fixed around the tail. Initially animal shows particular pulse level, when the pulse rate is within the normal range. 'STRAT' switch is put on and the recorder records the blood pressure as SBP (systolic blood pressure). DBP (Diastolic blood pressure) and MABP (mean arterial blood pressure), which is displayed on monitor. The pressure can be easily read from the pre-calibrated monitor. Once all the values are displayed the recorder is switched off and for next measurement. Some procedures are allowed once when sufficient pulse level is attained. [Table1, 2]

Table 1. Hypertension induced in normotensive rat

Comp.	Exp. Animal Albino (Wistar) Rat	After 1hour			After 3 hour		
		SBP	DBP	MABP	SBP	DBP	MABP
[1]	1	142	105	124	135	107	121
	2	141	102	121	139	103	121
	3	140	105	123	141	105	124
	4	143	101	122	140	110	125
	5	145	105	125	145	100	121
[2]	1	142	112	127	140	103	121
	2	140	110	125	139	107	123
	3	138	106	122	141	103	122
	4	132	110	121	143	105	124
	5	140	108	124	138	102	120
[3]	1	143	110	127	134	102	118
	2	138	107	128	143	101	121
	3	140	108	125	141	104	120
	4	144	111	126	143	112	116
	5	144	106	125	144	109	128
[4]	1	142	109	126	143	111	126
	2	140	102	123	140	100	120
	3	142	105	124	135	107	121
	4	141	102	121	139	103	121
	5	140	105	123	141	105	124
[5]	1	141	110	129	142	108	125

	2	138	105	125	139	107	123
	3	132	104	128	142	102	122
	4	142	103	123	140	102	121
	5	141	110	124	143	105	123
[6]	1	139	108	124	141	103	122
	2	142	113	128	142	104	123
	3	141	109	125	144	103	124
	4	144	114	128	141	102	121
	5	146	104	125	142	102	122
[7]	1	140	106	123	142	106	124
	2	141	114	128	142	104	123
	3	146	108	127	144	104	124
	4	148	114	130	144	102	123
	5	144	112	132	142	104	123
[8]	1	142	112	127	140	102	121
	2	144	116	130	141	101	122
	3	142	110	126	139	104	123
	4	146	106	126	144	104	124
	5	148	106	127	146	102	124
[9]	1	151	112	133	146	101	124
	2	144	114	129	142	102	121
	3	139	114	127	135	103	119
	4	142	106	124	140	102	123
	5	140	105	128	138	104	121
[10]	1	143	105	124	139	107	121
	2	141	101	126	143	102	120
	3	141	110	126	143	108	119
	4	142	102	125	141	105	121
	5	139	111	124	138	106	120
Control	**Losartan**	125	-	-	-	-	-

Table 2. Reduction in blood pressure (mm Hg) at a dose of 50 µgm/kg animal body weight

Comp.	**Exp. Animal Albino (Wistar) Rat**	**After 1hour**			**After 3 hour**		
		SBP	**DBP**	**MABP**	**SBP**	**DBP**	**MABP**
[1]	1	122	103	114	121	104	112
	2	120	101	111	120	102	111
	3	118	104	111	123	101	112
	4	120	102	111	125	102	113
	5	122	106	114	122	100	111
[2]	1	124	112	118	121	102	112
	2	126	105	116	127	101	114
	3	126	109	117	122	106	114
	4	124	103	115	125	101	113
	5	128	105	114	127	102	114
[3]	1	125	101	113	123	104	116
	2	132	104	118	127	107	117

	3	135	105	120	129	102	116
	4	123	103	113	124	103	114
	5	122	106	114	123	107	115
[4]	1	135	102	119	124	101	112
	2	136	101	118	122	104	113
	3	134	100	117	126	104	115
	4	122	102	112	122	100	111
	5	123	103	116	124	110	117
[5]	1	131	100	123	121	106	110
	2	129	103	124	122	100	111
	3	133	105	118	127	104	114
	4	123	104	114	125	104	111
	5	129	102	119	121	102	110
[6]	1	125	104	118	123	101	112
	2	128	105	116	128	102	115
	3	129	101	117	126	104	115
	4	128	102	115	126	104	115
	5	131	103	117	124	102	113
[7]	1	127	103	115	125	102	114
	2	124	104	114	128	101	113
	3	122	102	111	123	102	112
	4	124	103	111	125	102	113
	5	122	102	114	123	100	111
[8]	1	123	111	118	128	104	116
	2	127	105	116	126	105	115
	3	129	108	119	124	104	114
	4	122	112	117	122	103	112
	5	126	114	120	128	107	117
[9]	1	125	103	114	126	102	114
	2	127	104	116	124	105	114
	3	125	108	117	122	108	115
	4	124	105	115	125	106	116
	5	122	109	116	126	106	116
[10]	1	136	101	118	122	104	113
	2	134	100	117	126	104	115
	3	122	102	112	122	100	111
	4	123	103	116	124	110	117
	5	125	104	115	125	106	116
Control	**Losartan**	117	-	-	-	-	-

Invasive Method (Direct Method): Male albino wistar (150-250 gm) rats were used and housed at 24±1^{0}C room temperature. The rats were anaesthetized with sodium chloride 0.9% solution, Drug solution 10-µg/100ml, and Heparin 500 I.U.solution urethane hydrochloride 50% w/v solution 80 mg/kg i.p. To set up the instrument firstly the level of mercury in the left arm of manometer was adjusted to 90-100 mm of Hg (normal blood pressure of rat).this was done in steps of 10mm at a time and the physiogram so obtained was used as calibration graph for calculations. The Jugular vein and carotid artery were surgically cannulated for drug administration for recording the blood pressure respectively. The trachea was cannulated in order

to provide artificial respiration to rat during the experiment. By means of three way stop cock and a stainless steel needle at the end of P.E. tubing was attached to arterial cannula for B.P., Transducers and the Venus cannula to a syringe. Then both the cannulas were filled by heparinized saline before the administration. Arterial cannula was connected via transducer to physiograph recorder. Several baseline readings of systolic and diastolic pressures were recorded. The physiograph shows the reduction of the blood pressure with compare to losratan. Synthesized compounds were screened in presence of Angiotensin II induced hypertension (0.5 µg/kg i.v.) [Table 3, 4].

Table: 3 Blood Pressure values for synthesized compounds over duration of 90 minutes

Comp. No.	**Mean Arterial Pressure After**									
	0 min.	10 min.	20 min.	30 min.	40 min.	50 min.	60 min.	70 min.	80 min.	90 min.
Losartan	**165**	**160**	**154**	**150**	**145**	**137**	**130**	**126**	**122**	**116**
1	176	172	165	157	150	145	141	136	131	128
2	175	169	161	156	150	144	138	130	127	125
3	178	176	170	165	159	151	143	137	130	126
4	171	168	160	155	149	141	137	132	128	125
5	180	175	168	162	156	150	145	139	136	128
6	170	167	163	158	153	149	144	139	135	129
7	172	168	163	157	152	148	142	135	127	121
8	166	160	154	146	142	137	133	130	128	125
9	174	168	164	161	156	148	142	137	130	124
10	182	176	170	164	157	151	146	139	133	127

Table: 4 Antihypertensive Activity of synthesized compounds

Compound. No	**Minimum Blood pressure value(mm Hg)**	**Duration of hypertension effect(min.)**
Losratan	116	90
1	121	100
2	116	100
3	118	102
4	114	95
5	117	105
6	115	110
7	114	95
8	117	115
9	120	100
10	121	105

RESULTS AND DISCUSSION

4-chloro-1,2-phenylenediamine dihydrochloride (0.45 g, 2.5 mmol) in 5 ml of water was cooled to 0°C and treated with a solution of cyanogen bromide (0.60 ml, 5 M in acetonitrile, 3.0 mmol) and solid $NaHCO_3$ (0.41 mg, 4.9 mmol). The solution was stirred at ambient temperature for 40-42^o 4 hrs. N-[4-(6-Chloro -5-nitro-1H-benzoimidazol-2-yl)-phenyl]acetamide (1.12g, 0.01 mole) in absolute ethanol (30 mL) and various aromatic aldehydes (1.06 g, 0.01 mole) were taken and then an aqueous solution of KOH (2%, 5 mL) added to. it was dissolved in 20ml of DMF

(dimethyl formamide) and stirred vigorously with 1.5gm of potassium carbonate at 27^0C for four hour. To the resulting mixture 0.482gm of 4'bromomethylbiphenyl-2-carboxylic acid dissolved in 20 ml of DMF and then was added drop wise with dropping funnel in three hour the reaction was allowed to proceed for further 9 hours at room temperature and solvent removed under vacuum. The maximum activity has been observed with nitro group (Compound 4, 7,8 and 9). There are some sites in the receptor pocket, which can interact with the functional groups at position 5. Substituted benzimidazole nucleus coupled to carboxylbipheny methyl group has been designed, synthesized and evaluated for angiotensin II antagonism. Compound with amino group at 5-position and aromatic, aryl, alkyl compounds at 2- position have been found to be more potent than losratan.

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