Synthesis and biological evaluation of some novel substituted 2,6-dimethyl-1,4-dihydropyridine-3,5-dicarboxamides

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Synthesis and biological evaluation of some novel substituted 2,6-dimethyl -1,4-dihydropyridine-3,5-dicarboxamides

P Jaismy Jacob[1], Anoop Thomas[2], S Venkataraman[3], E Hareeshbabu[4] and S L Manju[1]

[1]Department of Chemistry, VIT University, Vellore, Tamil Nadu- 632 014, India
[2]Chemists College of Pharmaceutical Sciences and Research, Varikoli P O, Puthencruz, Kerala- 682 308, India
[3]Department of Pharmaceutical Chemistry, K. M. College of Pharmacy, Uthangudy, Madurai- 625 107, India
[4]Department of Pharmaceutical Chemistry, St. James College of Pharmaceutical Sciences, Chalakudy, Kerala -680 307, India

E-mail : slmanju@vit.ac.in

Abstract. A novel class of substituted 4-aryl-2,6-dimethyl-1,4-dihydropyridine-3,5-dicarboxamide has been synthesized by simple, modified Hantzsch condensation reaction using N-arylacetoacetamides, aryl aldehydes and ammonia. Characterisation of the newly synthesized compounds was carried out by spectral analysis (IR, 1H NMR and Mass Spectroscopy). Antimicrobial activity against *Staphylococcus aureus*, *Escherichia coli* and *Candida albicans* and antiulcer activity by aspirin induced and pyloric ligation ulcer model was studied. Results revealed that most of the compounds exhibit significant antimicrobial activity along with antiulcer activity. The compound **6i** 4-dimethylamino phenyl group at 4th position of 1,4-dihydropyridine had shown 65% ulcer protection at 10 mg/kg administration in male albino rats.

1. Introduction

Among a wide variety of heterocyclic molecules that have been developed for pharmaceutical importance, 1,4- dihydropyridine have played an important role. Molecules with 1,4- dihydropyridine nucleus received considerable attention as antimicrobial,[1] antitubercular,[2, 3] antiulcer,[4] anticonvulsant, analgesic and anti-inflammatory agents.[5] Because of its resemblance with nicotinamide adenine dinucleotide (NADH) and its biological activity as calcium channel blocker, this class of compounds achieved fast progress in medicinal chemistry. Acetyl choline, histamine and gastrin stimulate gastric acid secretion from parietal cells through Ca^{2+}. Yegen et al. reported that stress induced ulcers in rats could be prevented by calcium channel blockers.[6, 7] Nifedipin and several other calcium channel blockers are reported for their antiulcer activity.[8] Drugs commonly used for the treatment of peptic ulcers can be divided into four groups which include antacids, H_2 receptor antagonists, anticholinergics and proton pump inhibitors. Though these drugs are widely used clinically, they are associated with side effects such as dry mouth, giddiness, dizziness and constipation. Carbenoxolone, a glycerrhetic acid derivative used in the treatment of peptic ulcer is

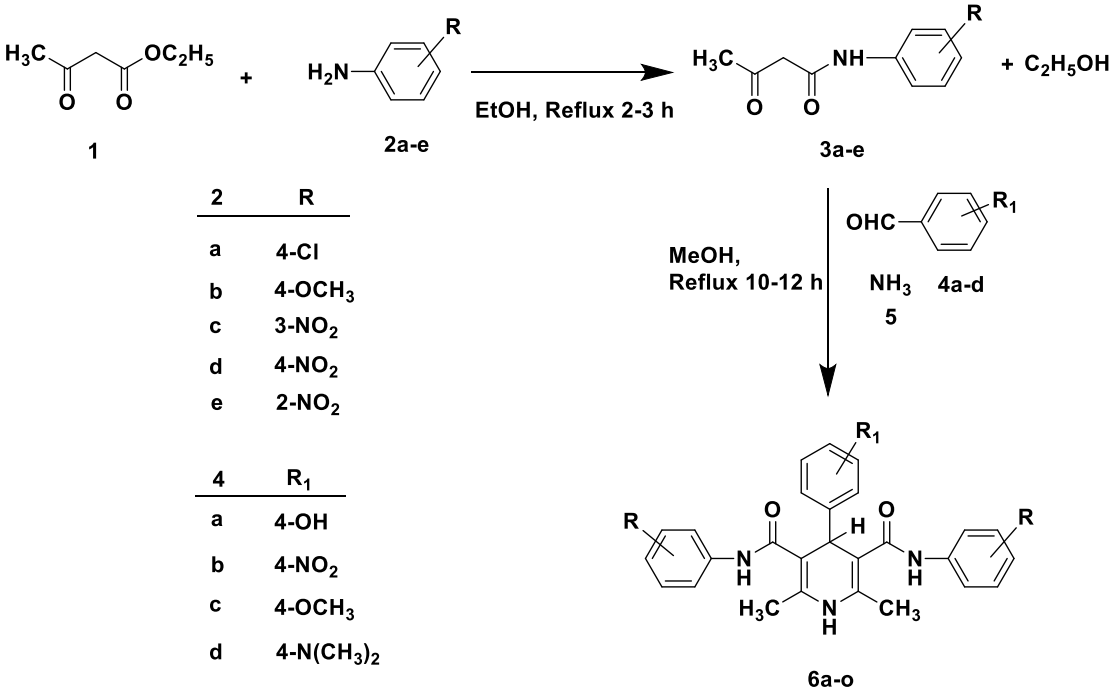
associated with salt retention and hyokalemia.[9] Hence, it is imperative to develop new chemical moieties with novel mechanism of action.

1,4- dihydropyridines (1,4-DHP) are mainly prepared by the well known Hantzch dihydropyrimidine synthesis, which involves 3 components such as aldehyde, active methylene compound (acetoacetic ester) and ammonia. Modifications in this reaction could be made by replacing β-keto ester with β-diketones, β-ketoacids, β-aminocrotononitrile, malonic acid esters and cyanoacetamide.[10] This research work was aimed to develop simple dihydropyridine derivatives which simultaneously exhibit antimicrobial and antiulcer activity.

2. Results and discussion

2.1. Chemistry

N-arylacetoacetamides (**3a-e**), the prime intermediates in the reaction, were synthesized by the condensation of ethylacetoacetate (1) with suitable aryl amines (2) in ethanol. Cyclocondensation of the N-arylacetoacetamides (**3a-e**) with various aldehydes (4a-d) and excess ammonia (5), afforded in single product in each case. The structures of 4-substituted-2,6-dimethyl-3,5-bis-N-(phenyl/substituted phenyl)-carbamoyl-1,4-dihydropyridines (**6a-o**) (Scheme 1), have been characterized by their spectral (IR, 1H NMR, Mass) and elemental analysis as presented in Table 1.

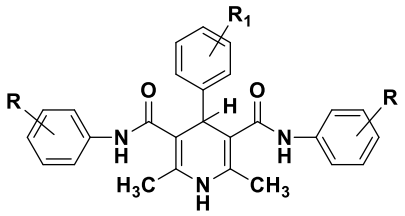


2	R
a	4-Cl
b	4-OCH_3
c	3-NO_2
d	4-NO_2
e	2-NO_2

4	R_1
a	4-OH
b	4-NO_2
c	4-OCH_3
d	4-$N(CH_3)_2$

Scheme 1. Synthesis of compounds **6a-o**. 1. Ethylacetoacetate, 2. Substituted amines, 3. N-arylacetoacetamide, 4. Substituted aldehydes, 5. Ammonia 25%, 6. 4- Substituted-4-aryl-2,6-dimethyl-1,4-dihydropyridine-3,5-dicarboxamide.

Table 1. Physical and analytical data of 1,4-DHPs **6a-o**.



6a-o

Compound	R	R_1	Mol. formula (Mol. wt)	m.p (ºC)	Yield (%)
6a	4-Cl	4-OH	$C_{27}H_{23}Cl_2N_3O_3$ 508.40	137-139	75
6b	4-Cl	4-NO_2	$C_{27}H_{22}Cl_2N_4O_4$ 537.40	144-146	80
6c	4-Cl	4-$N(CH_3)_2$	$C_{29}H_{28}Cl_2N_4O_2$ 535.47	139-141	87
6d	4-OCH_3	4-OH	$C_{29}H_{29}N_3O_5$ 499.57	180-182	85
6e	4-OCH_3	4-OCH_3	$C_{30}H_{31}N_3O_5$ 513.59	177-179	81
6f	4-OCH_3	4-$N(CH_3)_2$	$C_{31}H_{34}N_4O_4$ 526.64	189-191	92
6g	3-NO_2	4-OH	$C_{27}H_{23}N_5O_7$ 529.51	124-126	94
6h	3-NO_2	4- NO_2	$C27H_{22}N_6O_8$ 558.51	154-156	90
6i	3-NO_2	4-$N(CH_3)_2$	$C_{29}H_{28}N_6O_6$ 556.58	130-132	86
6j	4-NO_2	4-OH	$C_{27}H_{23}N_5O_7$ 529.51	180-182	81
6k	4-NO_2	4-OCH_3	$C_{28}H_{25}N_5O_7$ 543.54	168-170	92
6l	4-NO_2	4-$N(CH_3)_2$	$C_{29}H_{28}N_6O_6$ 556.58	164-166	88
6m	2-NO_2	4-OH	$C_{27}H_{23}N_5O_7$ 529.51	161-163	91
6n	2-NO_2	4- NO_2	$C_{27}H_{22}N_6O_8$ 558.51	142-144	89
6o	2-NO_2	4-$N(CH_3)_2$	$C_{29}H_{28}N_6O_6$ 556.58	158-160	71

2.2. Antibacterial screening

The *in vitro* antibacterial Screening of the synthesized compounds was carried out by the disc-diffusion method[11] in Muller-Hinton agar medium, at three different concentration, viz 25, 50 and 100 μg/ml using Gram positive *Staphylococcus aureus* (NCIM-5021) and Gram negative *Escherichia*

coli (NCIM-2911) strains. Reference used was Amikacin and DMSO as the control. Synthesized compounds were assessed in triplicate and results are presented in Table 2. Most of the compounds showed relatively higher activity towards Gram –ve organism. Compound 6m with 4-hydroxyphenyl group at 4th position had shown maximum activity against both Gram +ve and Gram –ve organisms.

Table 2. Antibacterial activity of 4-aryl/hetero aryl-3,5-bis-N-(phenyl/substituted phenyl)-carbamoyl-1,4-diydropyridines (6a-o).

Compounds	Zone of Inhibition in mm					
Code	*S. aureus*			*E. coli*		
	Concentration (µg/ml)			Concentration (µg/ml)		
	25	50	100	25	50	100
6a	7	8	9	8	10	11
6b	7	9	10	8	11	14
6c	8	10	12	12	14	17
6d	8	11	12	9	11	13
6e	7	8	9	8	12	14
6f	7	9	10	9	12	15
6g	10	12	13	10	12	14
6h	10	13	14	10	13	15
6i	13	15	16	14	16	18
6j	8	10	12	8	11	12
6k	8	10	11	14	17	19
6l	7	9	10	8	10	11
6m	13	16	19	16	19	22
6n	12	14	15	13	15	16
6o	8	10	11	10	12	14

Amikacin (30 μg/disc)	23	20

2.3. Antifungal Screening

The *in vitro* antifungal activity of the synthesized compounds was assessed by the disc-diffusion method in Sabouraud Dextrose Agar medium in triplicates at 25, 50 and 100 μg/ml using *Candida albicans* (NCIM 3100) strain. Standard used was Ketoconazole (10 μg/disc). After 24 hrs incubation at 37°C, zone of inhibition (mm) was measured. Antifungal activity of the synthesized compounds was depicted in the Table 3. Among the compounds 6m with 4-hydroxyphenyl group at 4th position exhibited significant antifungal activity comparable to the standard. Compounds 6b, 6c, 6e, 6f, 6g, 6i, and 6k exhibited moderate activity.

Table 3. Antifungal activity of various synthesized compounds against *Candida albicans*

Compounds	Zone of Inhibition in mm		
Code	*C. albicans*		
	Concentration in μg/ml		
	25	50	100
6a	9	11	12
6b	12	14	16
6c	14	17	19
6d	7	9	11
6e	12	14	15
6f	10	13	14
6g	14	17	19
6h	10	12	13
6i	9	12	14

6j	7	9	10
6k	13	15	16
6l	8	10	12
6m	17	19	22
6n	9	11	12
6o	8	11	13
Ketoconazole (10 µg/disc)	21		

2.4. Antiulcer activity

15 compounds were synthesized as substituted DHPs. Five differently substituted compounds were selected randomly viz. 6b, 6e, 6i, 6j and 6m for screening anti-ulcer activity in aspirin induced gastric ulceration in rat model.

2.4.1. Animals

Male albino rats (180-220 g) were selected. The animals were kept on a standard diet and water *ad libitum*. The animals were housed in group of six and accommodated to room condition for at least two days prior to the experiments. Food and water were freely available up to the experiments. The food was withdrawn 24 hour before ulcerogenic treatment, but free access to water was allowed. Synthesized molecules were evaluated for antiulcer activity against experimentally induced gastric ulcer models: aspirin plus pylorus ligation model in rats. The study was carried out after obtaining institutional animal ethical committee clearance.

2.4.2. Acute toxicity studies

The acute toxicity study was carried out by the method of Smith(1960) in wistar albino rats.

2.4.3. Anti-ulcer activity

The albino rats are weighing between 180-220g were divided into eight groups, each group contains six animals. The study was carried out for four days. 30 minutes after administration of the treated dose, the rats were treated with Acetyl salicylic acid (aspirin) 200mg /kg. This process was carried out for three days, on the third day after administration of drug the rats were subjected to fasting. After the fasting period the rats were anaesthetized by means of diethyl ether. The abdomen was opened and the pyloric end was ligated with a thread. [12, 13] Pyloric ligation was done after 60 minutes of sample administration. Group-I received 1% Carboxy methyl cellulose, CMC (1ml/kg, p.o.) acted as normal

control. Group-II received Acetyl salicylic acid, ASA (200 mg/kg, p.o.) acted as ulcer control. Group-III received Omeprazole (2 mg/kg, p.o) acted as standard control. The remaining groups received test compounds of 10 mg/kg, p.o 30 minutes prior to ASA administration. The rats were sacrificed four hours later by euthanization (CO_2 Chamber) and the oesophagi were clamped, the stomach was carefully exposed, opened along the greater curvature, gastric juice was collected after removing luminal contents. The gastric juice was centrifuged at 1000 rpm for 10 minutes. The total volume of gastric secretion, total acidity, and free acidity was estimated by titration method.

The gastric ulcer index was calculated according to the method of Gangly and Bhatnagar (1973). [14]

Mean ulcer score for each animal was expressed as ulcer index. The percentage of ulcer inhibition was determined as follows

$$\text{Protection of ulcer (\%)} = \frac{\text{Control mean ulcer index- test mean ulcer index}}{\text{Control mean ulcer index}} \times 100$$

Statistical analysis

The values were expressed as mean ± SEM. Statistical analysis was performed by one way analysis of variance (ANOVA) followed Newman kevel's multiple range test, probability value <p.0.01 was considered significant.

Table 4. Antiulcer Activity of Various Synthesized Compounds

Group	Treatment	Dose mg/kg	Total volume of gastric secretion (ml/100 gm)	Total acidity (meq/l/100g)	P^H	Ulcer score	% protection
I	Normal control	1 ml of 1%CMC	3.8± 0.55	418.44± 21.40	2.1± 0.24	0.3± 0.01	0.000
II	Ulcer control	200mg/kg ASA	5.2± 0.76^{*a}	493.53± 24.20^{*a}	1.5± 0.16^{*a}	2.0± 0.16^{*a}	0.000
III	Standard control	2mg/kg Omeprazole	2.5± 0.25	333.96± 20.12	4.2± 0.60	0.5± 0.10	75.00
IV	Treatment control	6b 10mg/kg drug	3.1± 0.34^{*b}	381.4± 29.45^{*b}	3.1± 0.34^{*b}	0.8± 0.15^{*b}	60.00
V	Treatment control	6e 10mg/kg drug	3.4± 0.26^{*b}	362.7± 26.02^{*b}	2.9± 0.30^{*b}	0.9± 0.18^{*b}	55.00
VI	Treatment control	6i 10mg/kg drug	3.0± 0.30^{*b}	368.4± 24.22^{*b}	3.2± 0.29^{*b}	0.7± 0.20^{*b}	65.00

VII	Treatment control	6j 10mg/kg drug	3.5± 0.22*b	342.1± 22.20*b	2.8± 0.39*b	0.9± 0.2*b	55.00
VIII	Treatment control	6m 10mg/kg drug	2.9± 0.20*b	321.7± 21.30*b	2.9± 0.27*b	0.72±0.12*b	64.00

*Values are expressed as Mean ± SEM

*a – Values are significantly different from Normal control group at P<0.01

*b -- Values are significantly different from ulcer control group at P<0.01

3. Conclusion

A new series of 1,4-dihydropyridine3,5-dicarboxamide derivatives **6a-o** were synthesized in high yields. These new 1,4-Dihydropyridines (1,4-DHPs) were evaluated for antibacterial, antifungal and antiulcer activities. Synthesized (1,4-DHPs) showed better activity towards Gram –ve bacteria than Gram +ve strains. The compound **6m** with 4-hydroxyphenyl substitution at 4th position of pyridine ring exhibited maximum sensitivity against both Gram +ve and Gram –ve bacteria. Most of the synthesized compounds displayed significant antifungal activity against *C. albicans*. Comparison of antibacterial and antifungal activities of compounds **6a-o** showed that **6m** possess potent antibacterial and antifungal activities. The compound **6m** exhibited better activity than the standard Ketoconazole at 100 μg/ml. The antiulcer activities have been performed by estimating the total volume of gastric secretion, total acidity, pH and ulcer index. Test compound **6i** with 4-dimethylamino phenyl group at 4th position of 1,4-DHPs has been found to be superior in antiulcer activity (65%) in comparison to other compounds in the series. Compounds **6m** (64%) and **6b** (60%) also showed significant ulcer protection compared to control group. The compound **6m** has emerged as a promising lead with potent antimicrobial and antiulcer activities. Hence this class of 1,4-DHPs afforded a special interest as it could simultaneously act as antimicrobial and antiulcer agent. Further studies could be carried out for the development of DHPs as dual acting antimicrobial and antiulcer agents.

4. Experimental

4.1. Chemistry

Melting points were determined using VEEGO-VMPT melting point apparatus in open capillaries and are uncorrected. IR spectra of the compounds were recorded on a Perkin Elmer FTIR spectrometer in KBr discs. ^{1}H NMR spectra were recorded on Bruker- Ultra shield FT-NMR (400 MHz) spectrometer in $CDCl_3$ or DMSO-d_6 with tetramethylsilane (TMS) as internal standard and the chemical shifts are reported as δ (ppm). Some representative molecules were studied for ^{13}C NMR spectra and were recorded in $CDCl_3$ or DMSO-d_6. Mass spectra were recorded on a GCMS Perkin Elmer instrument (70 eV). Thin layer chromatography was used to study the progression of the reaction and purity of the products on precoated silica gel 60 aluminium plates (Merck, Germany).

4.1.1. General procedure for the preparation of N-arylacetoacetamides(3a-e)

A mixture of equimolar quantities of ethylacetoacetate (1, 10 mmol) and aromatic amine (2, 10 mmol) were taken in a dry 250 ml RB flask and 20ml of ethanol was added and dissolved. The reaction mixture was refluxed for about 2-3 hrs. The solvent was removed under vacuum and the remaining was cooled. The precipitate was filtered, washed with cold water and dried. Purification of the crude product was done by recrystallization from ethanol.

4.1.2. General procedure for the preparation of 4-aryl-2,6-dimethyl -1,4-dihydropyridine-3,5-dicarboxamide (6a-m)

N-arylacetoacetamide (3a-e, 10 mmol) and substituted benzaldehyde (4a-d, 20 mmol) was dissolved in methanol and excess ammonia (25%) was added. Then it was stirred for 10 minutes and refluxed on a water bath for 10-12 hrs. Completion of the reaction was monitored by TLC. Methanol was removed under reduced pressure and cooled. The obtained product was filtered washed with methanol and dried. Further, it was purified by ethanol recrystallization or by column chromatography by petroleum ether –chloroform (3:1).

*4.1.2.1 N^3,N^5-bis(4-chlorophenyl)-4-(4-hydroxyphenyl)-2,6-dimethyl-1,4-dihydropyridine-3,5-dicarboxamide (**6a**)* (IR KBr, cm^{-1}): 3460 (O-H), 3262 (N-H), 1675 (C=O), 1548 (N-H bend, amide), 1286 (C-N), 816 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.56 (s, 6H, 2-CH_3), 4.72(s,1H, OH), 5.25 (s, 1H, H_4, DPH), 5.78 (s, 1H, NH-DHP), 6.85-8.16 (m, 12H, Ar-H), 9.84 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 21.43, 41.13, 109.2, 120.86, 123.61, 129.12, 131.26, 133.45, 135.95, 137.58, 148.12, 155.75, 165.38. m/z: 508[M^+]; Calculated mass: 508.40

*4.1.2.2 N^3,N^5-bis(4-chlorophenyl)-2,6-dimethyl-4-(4-nitrophenyl)-1,4-dihydropyridine-3,5-dicarboxamide (**6b**)* (IR KBr, cm^{-1}): 3248 (N-H), 1652 (C=O), 1539 (N-H bend, amide), 1290 (C-N), 810 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.52 (s, 6H, 2-CH_3), 5.20 (s, 1H, H_4, DPH), 5.82 (s, 1H, NH-DHP), 6.80-8.12 (m, 12H, Ar-H), 9.78 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 21.38, 41.18, 108.86, 121.18, 123.63, 129.20, 131.28, 133.46, 135.94, 137.52, 141.24,148.10, 165.40. m/z: 537 [M^+]; Calculated mass: 537.40

*4.1.2.3 N^3,N^5-bis(4-chlorophenyl)-4-(4-(dimethylamino)phenyl)-2,6-dimethyl-1,4-dihydropyridine-3,5-dicarboxamide (**6c**)* (IR KBr, cm^{-1}): 3342 (N-H), 1708 (C=O), 1560 (N-H bend, amide), 1278 (C-N), 790 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.54 (s, 6H, 2-CH_3), 3.12 (s, 6H N(CH_3), 5.22 (s, 1H, H_4, DPH), 5.92 (s, 1H, NH-DHP), 6.78-8.24 (m, 12H, Ar-H), 9.95 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 21.41, 41.12, 43.45, 109.2, 120.86, 123.61, 129.14, 131.25, 133.43, 136.18, 137.54, 147.82, 148.12, 166.21. m/z: 535 [M^+]; Calculated mass: 535.37

*4.1.2.4 4-(4-hydroxyphenyl)-N^3,N^5-bis(4-methoxyphenyl)-2,6-dimethyl-1,4-dihydropyridine-3,5-dicarboxamide (**6d**)* (IR KBr, cm^{-1}): 3446 (O-H), 3225 (N-H), 1689 (C=O), 1556 (N-H bend, amide), 1281 (C-N), 808 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.55 (s, 6H, 2-CH_3), 3.84 (s, 6H, OCH_3), 4.70 (s,1H, OH), 5.21 (s, 1H, H_4, DPH), 5.83 (s, 1H, NH-DHP), 6.82-8.18 (m, 12H, Ar-H), 9.84 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 21.46, 41.21, 57.35, 108.97, 120.81, 122.98, 129.24, 133.16, 134.48, 137.78, 138.82, 148.31, 155.78, 166.08. m/z: 499[M^+]; Calculated mass: 499.57

*4.1.2.5 N^3,N^5,4-tris(4-methoxyphenyl)-2,6-dimethyl-1,4-dihydropyridine-3,5-dicarboxamide (**6e**)* (IR KBr, cm^{-1}): 3316 (N-H), 1690 (C=O), 1542 (N-H bend, amide), 1276 (C-N), 788 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.45 (s, 6H, 2-CH_3), 3.84 (s, 6H, OCH_3), 3.91(s, 3H, OCH_3), 5.53 (s, 1H, H_4, DPH), 5.84 (s, 1H, NH-DHP), 7.19-7.86 (m, 12H, Ar-H), 9.79 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 20.21, 40.48, 55.51, 57.68, 108.82, 118.23, 121.06, 126.94, 131.72, 135.12, 137.29, 152.63, 159.38, 167.67. m/z: 513[M^+]; Calculated mass: 513.59

*4.1.2.6 4-(4-(dimethylamino)phenyl)-N^3,N^5-bis(4-methoxyphenyl)-2,6-dimethyl-1,4-dihydropyridine-3,5-dicarboxamide (**6f**)* (IR KBr, cm^{-1}): 3352 (N-H), 1698 (C=O), 1546 (N-H bend, amide), 1310(C-N), 794 (C-H aromatic);); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.50 (s, 6H, 2-CH_3), 3.10 (s, 6H N(CH_3), 3.82 (s, 6H, OCH_3), 5.23 (s, 1H, H_4, DPH), 5.91 (s, 1H, NH-DHP), 6.87-8.18 (m, 12H, Ar-H), 9.84 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 21.41, 40.12, 43.73, 55.58, 109.37, 120.21, 123.85, 128.38, 131.23, 133.26, 136.18, 142.82, 148.12, 155.48, 166.21. m/z: 526 [M^+]; Calculated mass: 526.64

*4.1.2.7 4-(4-hydroxyphenyl)-2,6-dimethyl-N^3,N^5-bis(3-nitrophenyl)-1,4-dihydropyridine-3,5-dicarboxamide (**6g**)* (IR KBr, cm^{-1}): 3465 (O-H), 3246 (N-H), 1695 (C=O), 1540 (N-H bend, amide), 1318 (C-N), 722 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.46 (s, 6H, 2-CH_3), 4.76 (s,1H, OH), 5.66 (s, 1H, H_4, DPH), 5.89 (s, 1H, NH-DHP), 7.32-8.14 (m, 12H, Ar-H), 9.95 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 20.34, 39.53, 106.28, 120.46, 123.18, 123.92, 128.25, 128.36, 133.65, 135.72, 147.68, 148.12, 155.75, 165.38. m/z: 529[M^+]; Calculated mass: 529.51

*4.1.2.8 2,6-dimethyl-N^3,N^5-bis(3-nitrophenyl)-4-(4-nitrophenyl)-1,4-dihydropyridine-3,5-dicarboxamide (**6h**)* (IR KBr, cm^{-1}) 3326 (N-H), 1689 (C=O), 1546 (N-H bend, amide), 1278 (C-N), 751 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.45 (s, 6H, 2-CH_3), 5.57 (s, 1H, H_4, DPH), 5.96 (s, 1H, NH-DHP), 6.92-8.16 (m, 12H, Ar-H), 9.98 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 20.12, 40.51, 106.28, 120.47, 122.85, 125.06, 128.32, 130.70, 132.18, 136.81, 146.73, 148.52, 154.58, 167.16. m/z: 558[M^+]; Calculated mass: 558.51

*4.1.2.9 4-(4-(dimethylamino)phenyl)-2,6-dimethyl-N^3,N^5-bis(3-nitrophenyl)-1,4-dihydropyridine-3,5-dicarboxamide (**6i**)* (IR KBr, cm^{-1}): 3312 (N-H), 1663 (C=O), 1542 (N-H bend, amide), 1353 (C-N), 691 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.32 (s, 6H, 2-CH_3), 3.10 (s, 6H N(CH_3), 5.24 (s, 1H, H_4, DPH), 5.90 (s, 1H, NH-DHP), 7.12-8.32 (m, 12H, Ar-H), 9.88 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 20.42, 41.16, 43.43, 109.14, 120.42, 123.63, 129.15, 131.22, 133.41, 136.20, 137.53, 147.80, 148.48, 167.2 m/z: 556 [M^+]; Calculated mass: 556.58

*4.1.2.10 4-(4-hydroxyphenyl)-2,6-dimethyl-N^3,N^5-bis(4-nitrophenyl)-1,4-dihydropyridine-3,5-dicarboxamide (**6j**)* (IR KBr, cm^{-1}): 3456 (O-H), 3320 (N-H), 1665 (C=O), 1547 (N-H bend, amide), 1298 (C-N), 788 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.41 (s, 6H, 2-CH_3), 4.74(s,1H, OH), 5.26 (s, 1H, H_4, DPH), 5.81 (s, 1H, NH-DHP), 7.06 -8.21 (m, 12H, Ar-H), 9.94 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 21.32, 41.32, 109.23, 116.76, 120.82, 123.32, 129.52, 135.56, 146.62, 148.22, 155.64, 167.44 m/z: 529[M^+]; Calculated mass: 529.51

*4.1.2.11 4-(4-methoxyphenyl)-2,6-dimethyl-N^3,N^5-bis(4-nitrophenyl)-1,4-dihydropyridine-3,5-dicarboxamide (**6k**)* (IR KBr, cm^{-1}): 3320 (N-H), 1686 (C=O), 1546 (N-H bend, amide), 1278 (C-N), 786 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.47 (s, 6H, 2-CH_3), 3.82 (s, 3H, OCH_3), 5.23 (s, 1H, H_4, DPH), 5.85 (s, 1H, NH-DHP), 6.96 -8.35 (m, 12H, Ar-H), 9.86 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 21.38, 41.26, 57.32, 108.86, 116.91, 120.78, 122.14, 134.48, 137.78, 148.31, 156.76, 167.28. m/z: 543[M^+]; Calculated mass: 543.54

*4.1.2.12 4-(4-(dimethylamino)phenyl)-2,6-dimethyl-N^3,N^5-bis(4-nitrophenyl)-1,4-dihydropyridine-3,5-dicarboxamide (**6l**)* (IR KBr, cm^{-1}): 3314 (N-H), 1672 (C=O), 1550 (N-H bend, amide), 1355 (C-N), 695 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.31 (s, 6H, 2-CH_3), 3.12 (s, 6H N(CH_3), 5.26 (s, 1H, H_4, DPH), 5.91 (s, 1H, NH-DHP), 6.93-8.12 (m, 12H, Ar-H), 9.83 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 20.38, 41.21, 43.45, 110.26, 120.39, 123.56, 129.61, 130.57, 133.02, 136.46, 138.50, 147.82, 148.14, 167.26 m/z: 556[M^+]; Calculated mass: 556.58

*4.1.2.13 4-(4-hydroxyphenyl)-2,6-dimethyl-N^3,N^5-bis(2-nitrophenyl)-1,4-dihydropyridine-3,5-dicarboxamide (**6m**)* (IR KBr, cm^{-1}): 3460 (O-H), 3322 (N-H), 1660 (C=O), 1542 (N-H bend, amide), 1300 (C-N), 786 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.43 (s, 6H, 2-CH_3), 4.75(s,1H, OH), 5.25 (s, 1H, H_4, DPH), 5.83 (s, 1H, NH-DHP), 6.96 -8.32 (m, 12H, Ar-H), 9.94 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 21.34, 41.30, 109.17, 116.62, 120.85, 122.63, 129.45, 135.41, 146.23, 148.42, 155.52, 167.47 m/z: 529[M^+]; Calculated mass: 529.51

*4.1.2.14 2,6-dimethyl-N^3,N^5-bis(2-nitrophenyl)-4-(4-nitrophenyl)-1,4-dihydropyridine-3,5-dicarboxamide (**6n**)* (IR KBr, cm^{-1}): 3324 (N-H), 1681 (C=O), 1542 (N-H bend, amide), 1282 (C-N), 782 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.40 (s, 6H, 2-CH_3), 5.56 (s, 1H, H_4, DPH), 5.94 (s, 1H, NH-DHP), 6.91-8.36 (m, 12H, Ar-H), 9.95 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 20.13, 40.53,

106.26, 120.45, 122.84, 126.03, 129.31, 130.79, 132.64, 137.82, 146.83, 148.60, 155.52, 167.28. m/z: 558[M^+]; Calculated mass: 558.51

*4.1.2.15 4-(4-(dimethylamino)phenyl)-2,6-dimethyl-N^3,N^5-bis(2-nitrophenyl)-1,4-dihydropyridine-3,5-dicarboxamide (**6o**)* (IR KBr, cm^{-1}): 3320 (N-H), 1665 (C=O), 1536 (N-H bend, amide), 1356 (C-N), 694 (C-H aromatic); ^{1}H NMR ($CDCl_3$) δ (ppm) 2.34 (s, 6H, 2-CH_3), 3.26 (s, 6H N(CH_3), 5.25 (s, 1H, H_4, DPH), 5.85 (s, 1H, NH-DHP), 7.06-8.37 (m, 12H, Ar-H), 9.85 (s, 2H, 2 CONH); ^{13}C NMR δ (ppm): 20.46, 41.15, 43.38, 109.26, 121.38, 123. 86, 128.64, 131.46, 134.38, 136.68, 138.04, 147.64, 149.32, 167.32 m/z: 556[M^+]; Calculated mass: 556.58

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