Research Article

Synthesis Bioactivities and Non-linear Optical Behavior of Angularly and Linearly Fused Pyridazino Quinoline Derivatives

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ABSTRACT

Angularly and linearly fused pyridazino quinolines has been reported via the cyclisation of hydrazino quinoline with chloroacetyl chloride. The structure was characterized by IR and NMR studies. The synthesized chemical compounds were applied against 2 gram-positive bacteria and 2 gram-negative bacteria by the well diffusion method. The entire synthesized compounds exhibited different degrees of antimicrobial activity at concentrations between 20–100 µg/disc against the test organisms. Antioxidant studies were also performed against DPPH radical and the results were attractive. *In vivo* anti-inflammatory and analgesic screening was also done for selected drug and the results were desirable. Quantum chemical calculations were performed using the DFT B3LYP method in the 6-31G (d, p) basis. Non-linear optical behavior of the compounds was analyzed by with first hyperpolarisability. The non-linear optical properties of solution of compound in DMSO solvent has been investigated using Z-scan technique with femto second. The calculated value of non-linear absorption coefficient displayed the non-linearity of the molecule. The *in silico* ad instrumental results showed that the compounds are attractive molecules for future application in non-linear optics.

Key words: Analgesic, Antiinflammatory, Carrageenan, Chloroacetyl chloride, Pyridazine, S. *aureus*.

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INTRODUCTION

Pyridazine derivatives are very attractive from the pharmaceutical point of view.[1] They have been reported to exhibit a variety of biological features ranging from anticancer and antituberculosis[2] to antibacterial, antimicrobial[3] and various other kind of biological activities.[4] Some pyridazines are used in the treatment of Parkinson's, Alzheimer's, and other neurodegenerative diseases.[5] Recently, pyridazines have been considered by GlaxoSmithKline as one of the "most developable" hetero aromatic rings for drug design.[6]

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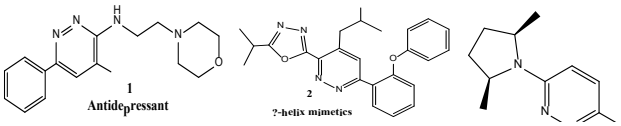
Pyridazines were recognized as selective GABA-A receptor antagonists, such as minaprine 1.[7] Volonterio *et al.* developed the synthesis of pyridazine-based scaffolds such as 2 to target protein/protein interaction as α-helix mimetics.[8] 3-Amino-6-aryl-pyridazines have been considered as an interesting pharmacophore in drug discovery. Some compounds show biological activity ranging from obesity[9] or neurodegenerative diseases[10] to inflammatory pain such as the selective CB2 agonist 3.[11]



MATERIALS AND METHODS

Pharmacological importance of pyridazinones intended us to synthesize the novel pyridazinones via their hydrazine intermediates. The precursor for the synthesis of pyridazinones and their derivatives namely 2-hydroxy-

4-phenyl and 4-hydroxy-2-phenyl quinoline and thus corresponding chloro derivatives were also prepared. The chloro quinoline thus obtained was converted in to corresponding hydrazine derivative by treating it with equal moles of hydrazine hydrate. The aimed compounds were then obtained by refluxing equal moles of hydrazino quinoline and chloro acetyl chloride in DMSO. The progress of the reaction was monitored by TLC. After the completion of the reaction, the reaction mixture was poured into ice and separated with benzene and the solvent was removed under reduced pressure. The product separated was column chromatographed over silica gel with petroleum ether: ethyl acetate (94:6) as eluant and recrystallised from ethanol

Preparation of substituted pyridazino quinolines

1. 8-methyl-5-phenyl-1,2-dihydropyridazino[3,4-*b*] quinolin-3(4*H*)-one 4b:- 2-hydrazino-7-methyl-4- phenyl quinoline: 2g (8.1mmoles), Chloroacetyl chloride: 0.6mL (8.1mmoles), Benzene: 20mL , *N,N*-dimethyl aniline: 2drops, Refluxing time: 15 h, Yield: 56%, Melting point: 148°C,IR (cm^{-1}): 2930(NH), 2855(NH_2), 1721 (C=O), 1626(CN). ^{1}H-NMR (DMSO) (ppm): δ1.42(*s*,3H,CH_3), δ8.9(*s*,1H,NH), δ5.38(*s*,1H, NH), δ 4.1(*s*,H,OH), δ 6.8-7.3(*m*,10H,Ar-H). ^{13}C-NMR (DMSO) (ppm): δ 162(C=O), δ46 (CH_3-C), δ98-δ 140(Ar-C & CH_2).

2. 7-methyl-5-phenyl-1,2-dihydropyridazino[3,4-*b*] quinolin-3(4*H*)-one 4c:- 2-hydrazino-6-methyl-4- phenyl quinoline: 2.2g (8.9mmoles), Chloroacetyl chloride: 0.7mL (8.9mmoles), Benzene: 20mL , *N,N*-dimethyl aniline: 2drops, Refluxing time: 9.5 h, Yield: 53.6%, Melting point: 153°C, IR (cm^{-1}): 2986(NH), 3110(NH), 1517 (C=N), 1722(C=O).

^{1}H-NMR (DMSO) (ppm) : δ1.8(*s*,3H,CH_3), δ8.1(*s*,1H,NH), δ 5.8(*s*,1H,NH), δ 6.7-7.2(*m*,10H,Ar-H,CH_2).^{13}C-NMR (DMSO) (ppm): δ161(C=O), δ47 (CH_3-C), δ78-δ 136(Ar-C), δ 76(CH_2)

3. 9-methyl-5-phenyl-1,2-dihydropyridazino[3,4-*b*] quinolin-3(4*H*)-one 4d:- 2-hydrazino-8-methyl-4-phenyl quinoline: 1.8g (7.2mmoles), Chloroacetyl chloride: 0.6mL (7.2mmoles), Benzene: 20mL , *N,N*-dimethyl aniline: 2drops, Refluxing time: 9 h, Yield: 58%, Melting point: 189°C, IR (cm^{-1}) : 2920(NH), 2855(NH), 1600 (C=N), 1729(C=O).

4. 8-chloro-5-phenyl-1,2-dihydropyridazino[3,4-*b*] quinolin-3(4*H*)-one 4e:- 7-chloro-2-hydrazino-4- phenyl quinoline: 1.3g (4.9mmoles), Chloroacetyl chloride: 0.4mL (4.9mmoles), Benzene: 20mL , *N,N*-dimethyl aniline: 2drops, Refluxing time: 14.5 h, Yield: 56%, Melting point: 200°C, IR (cm^{-1}): 2929(NH), 2885(NH), 1563 (C=N), 1710(C=O).

5. 9-chloro-5-phenyl-1,2-dihydropyridazino[3,4-*b*] quinolin-3(4*H*)-one 4 f:- 8-chloro-2-hydrazino-4- phenyl quinoline: 1.32g (4.9mmoles), Chloroacetyl chloride: 0.4mL (4.9mmoles), Benzene: 20mL , *N,N*-dimethyl aniline: 2 drops, Refluxing time: 12h, Yield: 59.3%, Melting point: 202°C, IR (cm^{-1}): 2972(NH), 2910(NH), 1599 (C=N), 1698(C=O).

6. 7-chloro-5-phenyl-1,2-dihydropyridazino[3,4-*b*] quinolin-3(4*H*)-one 4g:- 6-chloro-2-hydrazino-4- phenyl quinoline: 1.5g (5.6mmoles)), Chloroacetyl chloride: 0.4mL (5.6mmoles), Benzene: 20mL , *N,N*-dimethyl aniline: 2drops, Refluxing time: 10h, Yield: 57.4%, Melting point: 198°C, IR (cm^{-1}): 2977(NH), 2935(NH), 1620 (C=N), 1724(C=O).

^{1}H-NMR (DMSO) (ppm): δ8.6(*s*, 1H,NH), δ 9.3(*s*,1H,NH), δ 6.3-7.8 (*m*,10H,Ar-H,CH_2).

^{13}C-NMR (DMSO) (ppm): δ164(C=O), δ78-δ 98-142(Ar-C, CH_2).

7. 7-methyl-5-phenyl-1,2-dihydropyridazino[4,3-*c*] quinolin-3(4*H*)-one 7b:- 4-hydrazino-8-methyl-2-phenyl quinoline: 1.5g (6mmoles), Chloroacetyl chloride: 0.5mL (6moles), Benzene: 20mL , *N,N*-dimethyl aniline: 2drops, Refluxing time: 9.5h, Yield: 61.26%, Melting point: 200°C, IR (cm^{-1}) : 2930(NH), 2859(NH), 1640 (C=N), 1721(C=O).

^{1}H-NMR (DMSO) (ppm) : δ1.6 (*s*,3H,CH_3), δ4.3,5.3(*bs*,OH, NHC=O↔N=C-OH), δ 8.9(*s*,1H,NH), δ 6.8-7.9(*m*,10H,Ar-H& CH_2).^{13}C-NMR (DMSO) (ppm) : δ158(C=O), δ28 (CH_3-C), δ144 (CN), δ78-δ142 (Ar-C and CH_2).

8. 9-methyl-5-phenyl-1,2-dihydropyridazino[4,3-*c*] quinolin-3(4*H*)-one 7c:- 4-hydrazino-6-methyl-2- phenyl quinoline: 1g (4.05mmoles), Chloroacetyl chloride: 0.5mL (6moles), Benzene: 20mL , *N,N*-dimethyl aniline: 2drops, Refluxing time: 10.5h, Yield: 63.89%, Melting point: 203°C, IR (cm^{-1}) :2930(NH), 2859(NH), 1550 (C=N), 1739(C=O).

^{1}H-NMR (DMSO) (ppm) : δ1.6 (*s*,3H,CH_3), δ8.4(*s*,1H,NH), δ 8.9(*s*,1H, NH), δ 6.8-7.9(*m*,10H,Ar-H& CH_2).^{13}C-NMR (DMSO) (ppm) : δ171(C=O), δ48 (CH_3-C), δ159(CN), δ121-δ146 (Ar-C), δ 82(CH_2).

9. 5-phenyl-1,2-dihydropyridazino[4,3-*c*]quinolin-3(4*H*)-one 7d:- 4-hydrazino -2- phenyl quinoline: 1g(4.3mmoles), Chloroacetyl chloride: 0.5mL (6moles), Benzene: 20mL, *N,N*-dimethyl aniline: 2drops, Refluxing time: 16h, Yield: 65%, Melting point: 189°C, IR (cm^{-1}) : 2921(NH), 2852(NH), 1584 (C=N), 1736(C=O).

10. 7-chloro-5-phenyl-1,2-dihydropyridazino[4,3-*c*] quinolin-3(4*H*)-one 7e:- 8-chloro - 4-hydrazino -2-phenyl quinoline: 1.35g (5.0mmoles), Chloroacetyl chloride: 0.4mL(5.0moles)), Benzene: 20mL, *N,N*-

dimethyl aniline: 2drops, Refluxing time: 14h, Yield: 62%, Melting point: 220°C,IR (cm^{-1}) :2891(NH), 2835(NH), 1627 (C=N), 1725(C=O).

11. 8-chloro-5-phenyl-1,2-dihydropyridazino[4,3-*c*] quinolin-3(4*H*)-one 7f:- 7-chloro- 4-hydrazino -2-phenyl quinoline: 1g (3.7mmoles), Chloroacetyl chloride: 0.3mL (3.7mmoles), Benzene: 20mL , *N,N*-dimethyl aniline: 2 drops, Refluxing time: 12.5h, Yield: 64.95%, Melting point: 227°C,IR (cm^{-1}): 2970(NH), 2926(NH), 1520 (C=N), 1656(C=O).^{1}H-NMR (DMSO) (ppm) : δ4.1 (*bs*, OH, NHC=O↔N=C-OH), δ 8.6(*s*,1H,NH), δ 6.8-7.3(*m*,10H,Ar-H& CH_2). ^{13}C-NMR (DMSO) (ppm): δ15(C=O), δ168 (CN), δ144 (CN), δ97-δ145 (Ar-C and CH_2).

12. 9-chloro-5-phenyl-1,2-dihydropyridazino[4,3-*c*] quinolin-3(4*H*)-one 7g:- 6-chloro- 4-hydrazino -2-phenyl quinoline: 1g (3.73mmoles), Chloroacetyl chloride: 0.3mL (3.73mmoles), Benzene: 20mL , *N,N*-dimethyl aniline: 2drops, Refluxing time: 1h, 0Yield: 66%, Melting point: 226°C

Pharmacological Screening

Then we attempted to compile the medicinal chemistry aspects of pyridazino quinoline derivatives not yet reported so far.

In vitro antibacterial *(S. aureus, S. pyogens, E. coli, Pseudomonas Sp.)*, antioxidant behaviour were determined. The *in-vivo* anti-inflammatory and analgesic studies were carried out for one of them by considering the purity and availability of the drug.

Antimicrobial Studies

Antibacterial studies were carried out with gram positive (*S.aureus, S. pyogens*) and gram negative (*E. coli, Pseudomonas* sp) bacteria by agar well diffusion method.[12] The sizes of the zone of inhibition were measured in millimeter and recoded. A series of standard antimicrobial drugs (common commercial antibiotics) were screened in par to compare the potency of the tested compounds.

Antioxidant Studies

Antioxidant studies were carried out for the compounds using DPPH radical scavenging activity method.[13] The percentage inhibition was calculated.

In vivo Studies

In vivo studies are those in which the effect of various biological systems is tested on living organisms or cells usually in animals. Balb/c mice (20–25 g) were used in the study. They were purchased from Sri Venkateswara Enterprises, Bengaluru, Suppliers of Laboratory animal. All the animal experiments were done as per the instructions prescribed by the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Ministry of Environment and Forest, Government of India, and implemented through the Institutional Animal Ethical Committee of the Research Centre. The drugs used is 5-phenyl-1,2-dihydropyridazino[4,3-c]quinolin-3(4*H*)-one 7d**.**

Antiflammatory activity

In vivo antiflammatory activity of selected compounds were done using carrageenan induced acute inflammatory model. The percentages of inhibition were calculated.[14] To study the effect of drug on inflammation we have chosen the third hour percentage inhibition as the inflammatory effect is maximum at its third hour of diclofenac.

Analgesic (Antinociceptive) Activity

Analgesic activity of selected compounds was screened by using acetic acid induced writhing model[14] with three groups of mice five animals in each group.

DFT studies for calculating NLO property

All calculations were done by using Gaussian 09W package of programs. Gaussian 09W[15] has already proved to be an important tool predicting molecular structures, molecular origins of NLO properties. The geometries of the compounds were optimized by Becke's three parameter hybrid functional for exchange combined with the correlation functional using 6-31G (d,p) basis set. Thus the NLO property of the material is theoretically calculated using mean polarizability (α_0), the total static dipole moment (μ) and the first order hyperpolarisability (β_0) with respect to *x, y, z* components using the equation given below and tabulated in Table 7.

$$\mu = (\mu x^2 + \mu y^2 + \mu z^2)^{1/2}$$

$$\alpha_0 = 1/3\ (\alpha_{XX} + \alpha_{YY} + \alpha_{ZZ})$$

$$\beta_0 = (\beta x^2 + \beta y^2 + \beta z^2)^{1/2}$$

The results obtained from the calculations were tabulated and the values were compared with that of urea (0.11x10-30esu),[16] since it is one of the molecules used in the study of the NLO properties of the molecules. And the values obtained for urea was used as the threshold value for the compounds.Gaussian outputs are reported in atomic units, so the calculated values were converted to esu [α (1au= $0.1482x10^{-24}$ esu) β (1au=$8.639x10^{-30}$esu)].

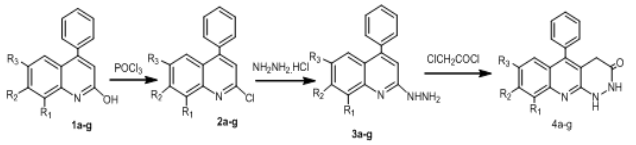
Z Scan technique

Z-scan technique was introduced by Mansoor Sheik-Bahae *et al.* in 1989.[17] The basic phenomena behind this are the optical Kerr effect.[18]

The Z-scan measurements in the ns regime were done using the standard open aperture Z-scan technique developed by M. Sheik Bahae *et al.*[17]

RESULTS AND DISCUSSION

Linearly fused pyridazinones were synthesised according to Scheme 1.



a)	R_1= R_2= R_3= H	b)	R_1= R_3=H, R_2= CH_3	c)	R_1= R_2= H, R_3= CH_3
d)	R_1=CH_3, R_2= R_3=H	e)	R_1= R_3=H, R_2=Cl	f)	R_1=Cl, R_2=R_3= H
g)	R_1= R_2=H, R_3= Cl				

Scheme 1

Initially, 2-chloro-4-pvhenyl quinoline is prepared by the reaction between 2-hydroxy-4- phenyl quinoline and electrophilic reagent phosphorus oxy chloride by the substitution of hydroxyl group with chlorine. The chloro derivative was then converted in to its hydrazino derivatives by the reaction with hydrazine hydrate. 5-phenyl-1,2-dihydropyridazino[3,4-*b*]quinolin-3(4*H*)-one was obtained by the reaction of 2-hydrazino-4-phenyl quinoline with chloroacetyl chloride in presence of DMA. All the conversions were confirmed with spectra.

IR υ(cm^{-1}) spectrum Figure 1 of 2-hydrazino-4-phenyl quinoline showed absorption peak at 2924 cm^{-1} due to NH stretching, and at 3055cm^{-1} due to NH_2. 1597cm^{-1} for C=N.

^{1}H-NMR (DMSO) (ppm) Figure 2 spectrum of the compound showed absorption of a one proton signal at δ 8.5 indicate the presence of NH_2 group, peak at δ 9.1 for NH proton, δ 6.9-7.7(m,10H,Ar-H).

Aromatic carbon atoms showed peaks from δ 65-148 in^{13}C-NMR ($CDCl_3$) (ppm) Figure 3 spectrum of the compound 3(a).

Formation of 5-phenyl-1,2-dihydropyridazino[3,4-*b*] quinolin-3(4*H*)-one was obtained by analyzing the following spectra.

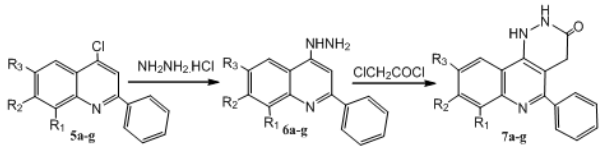
IR (cm^{-1}) Figure 4 spectrum of the compound showed absorption peak at 2929, 2850(NH), 1721 for carbonyl group and peak at 1599 cm^{-1} indicates the presence of CN.

Here disappearance of peak at 3055cm^{-1} due to NH_2 and appearance of peak at 2850cm^{-1} pinpointing the conversion.

^{1}H-NMR (DMSO) (ppm) Figure 5 spectrum of the compound showed t absorption at δ 8.9 indicating the presence of NH δ4.3, δ5.5 (bs,OH, NHC=O↔N=C-OH) aromatic protons and CH_2 protons of pyridazinone ring appeared at δ 6.9-7.8 as a multiplet.

^{13}C-NMR ($CDCl_3$) (ppm) Figure 6 spectrum of the compound 4a showed absorption at δ 167(C=O), δ30(C-4), δ90-150(C_5, C_6, C_7, C_8, C_9, C_{10}, C_{11}, C_{12}, C13, C_{14}, C_{15}, C_{16}, C_{17}, C_{18}, C_{19} aromatic carbon).

Then angularly fused pyridazinones were synthesized from its precursor 4-hydrazino2-phenyl quinoline derivatives depicted in Scheme 2.



a)	R_1= R_3=H, R_2=CH_3	b)	R_2= R_3=H, R_1=CH_3	c)	R_1= R_2= H, R_3= CH_3
d)	R_1= R_2= R_3=H	e)	R_2= R_3=H, R_1=Cl	f)	R_2=Cl, R_1=R_3=H
g)	R_1= R_2=H, R_3=Cl				

Scheme 2

8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-*c*] quinolin-3(4*H*)-one was obtained by refluxing 4-hydrazino-7-methyl-2-phenyl-quinoline 1g (4.03mmoles) and chloroacetyl chloride 0.3mL (4.03mmoles)in 20 mL of DMSO for 18 hrs. Formation of 4-hydrazino- 7-methyl-2-phenyl quinoline was confirmed with spectral details. IR (cm^{-1}) Figure 7 spectrum of the compound 6a showed absorption peak at 3120 (NH), 2984(NH_2), 1602(C=N).

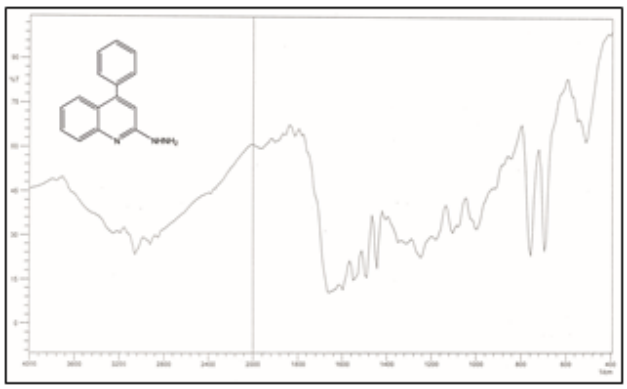


Figure 1: IR spectrum of 2-hydrazino-4-phenylquinoline 3a.

^{1}H-NMR (DMSO) (ppm) Figure 8 spectrum of the compound 6a showed absorption at δ2.1(s,3H,CH_3), δ9 (s,H, NH), δ8.7 for NH_2 protons, δ 6.7-7.4(m,9H,Ar-H).
^{13}C-NMR ($CDCl_3$) (ppm) Figure 9 spectrum of the compound 6a showed absorption at δ 182 is assigned for CN carbon atoms, δ21(CH_3), δ120-139 indicates the presence of aromatic carbon atoms.
Further the cyclisation was established by IR and NMR spectrum.
IR (cm^{-1}) Figure 10 spectrum of 8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-*c*]quinolin-3(4*H*)-one 7a showed absorption peak at 2923 for (NH), 2854(NH), 1677cm^{-1} for (C=O) group of pyridazinone ring, 1546(C=N).
^{1}H-NMR (DMSO) (ppm) Figure 11 spectrum of the compound 7a showed absorption at δ1.8(s,3H,CH_3), δ8.7(s, H, NH), δ4.5,5.5(*bs*, OH, NHC=O↔N=C-OH), δ6.9-8 (m,10H, Ar-H,CH_2 of pyridazinone ring).
^{13}C NMR ($CDCl_3$) (ppm) Figure 12 spectrum of the compound 7a showed absorption at δ167 (C=O), δ30(C-4), peak at δ21 indicates the presence of methyl carbon atom, δ90-150 isfor CN.
Figure 12: ^{13}C-NMR spectrum of 8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-c]quinolin-3(4H)-one 7a.

The behavior of the synthesized compounds towards the bacteria and fungi has also been studied. The DPPH radical scavenging activity was also studied by *in vitro*. Apart from this *in vivo* anti-inflammatory, analgesic studies were done for selected compound.

Pharmacological Activities

Antimicrobial activity

Antimicrobial activity of substituted 5-phenyl-1,2-dihydropyridazino[3,4-*b*]quinolin-3(4*H*)-one and 5-phenyl-1,2-dihydropyridazino[4,3-*c*]quinolin-3(4*H*)-one were tested with *S. aureus* and *S. pyogens* as gram positive and *E. coli* and *Pseudomonas Sp.* as gram negative bacterial stains. The results were tabulated and also discussed by the help of chart also.
The Table 1 and Figure 13 shows that antibacterial activity of pyridazino quinolones is remarkable. Against *s.aureus*, compound 5-phenyl-1,2-dihydropyridazino[4,3-*c*] quinolin-3(4*H*)-one 7d showed a maximum inhibition. Against *s. pyogens* almost all compounds showed a notable inhibition, and 5-phenyl-1,2-dihydropyridazino[4,3-*c*] quinolin-3(4*H*)-one 7d showed the maximum value.

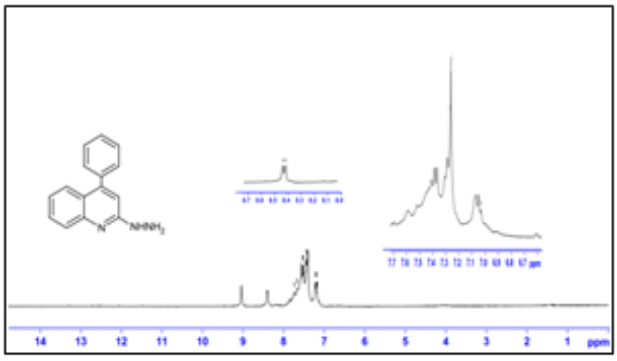


Figure 2: ^{1}H-NMR spectrum of 2-hydrazino-4-phenylquinoline 3a.

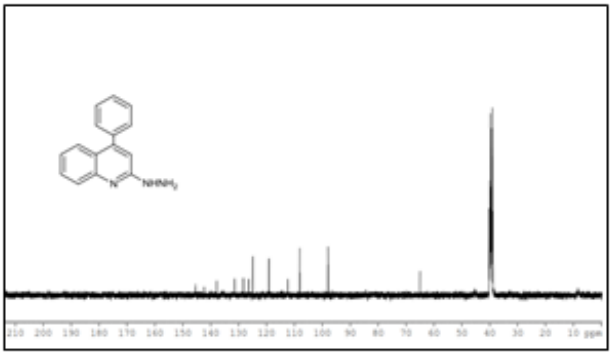


Figure 3: ^{13}C-NMR spectrum of 2-hydrazino-4-phenylquinoline 3a.

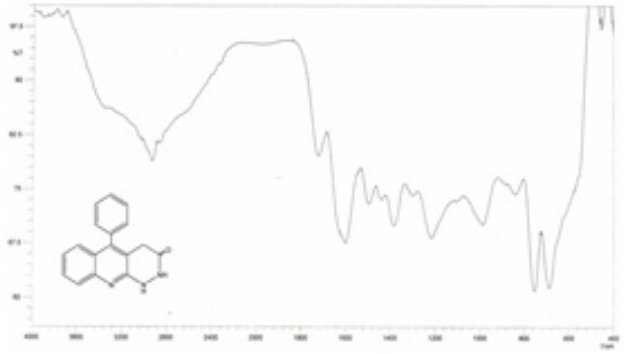


Figure 4: IR spectrum of 5-phenyl-1,2-dihydropyridazino[3,4-b]quinolin-3(4H)-one 4a.

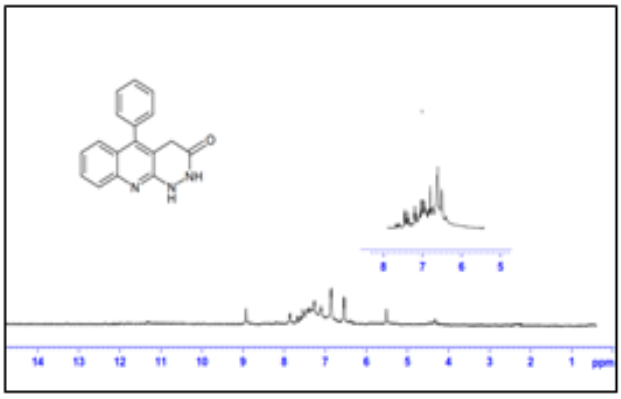


Figure 5: ^{1}H-NMR spectrum 5-phenyl-1,2-dihydropyridazino[3,4-b]quinolin-3(4H)-one 4a.

Even at 0.75µg/mL the inhibition is comparable with the higher concentration.

For *E. coli* Table 2compound 4f exhibited the highest activity. It supports 7b can act as a good inhibitor against *p. arogonosa.*

MIC of the substituted pyridazino quinolines was also screened by broth dilution method.[19] When we examine the MIC value of pyridazino quinolines all the compounds suppressed the growth of *S. aureus* bacteria at the concentration of 0.5µg/mL but inhibit the growth of *S. pyogens* in the concentration of 0.25 µg/mL itself. But gram negative bacteria need more drug concentration to stop its growth.

Antioxidant activity

Antioxidants neutralize the effect of free radicals, substances that damage the body's cells and have a role in disease prevention. It can be achieved by donating

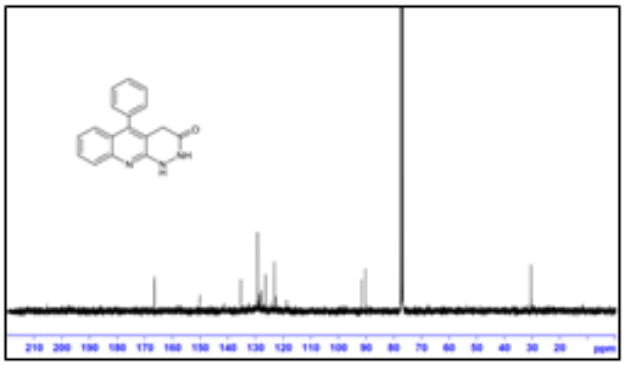


Figure 6: ^{13}C-NMR spectrum of 5-phenyl-1,2-dihydropyridazino[3,4-b]quinolin-3(4H)-one 4a.

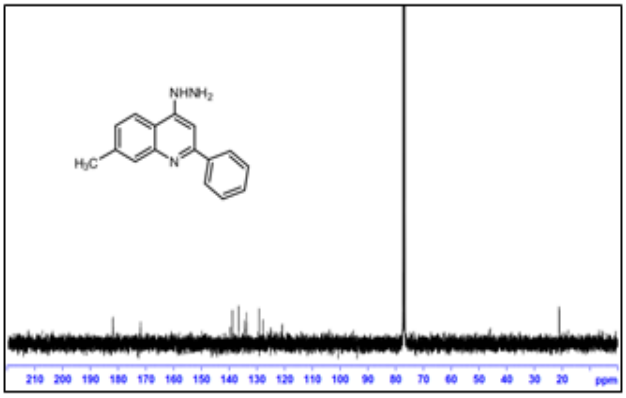


Figure 9: ^{13}C-NMR spectrum of 4-hydrazino- 7-methyl-2-phenyl- quinoline 6(a).

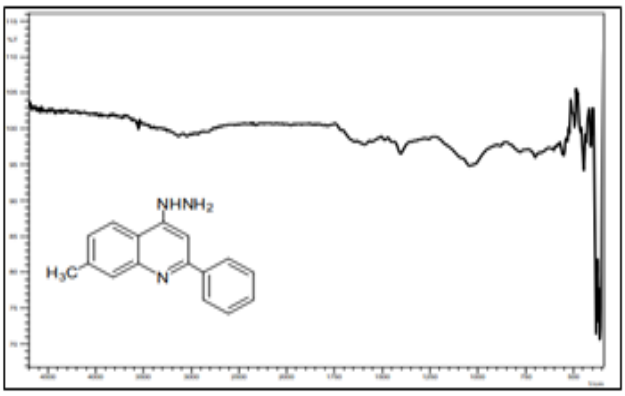


Figure 7: IR spectrum of 4-hydrazino-7-methyl-2-phenyl-quinoline 6a.

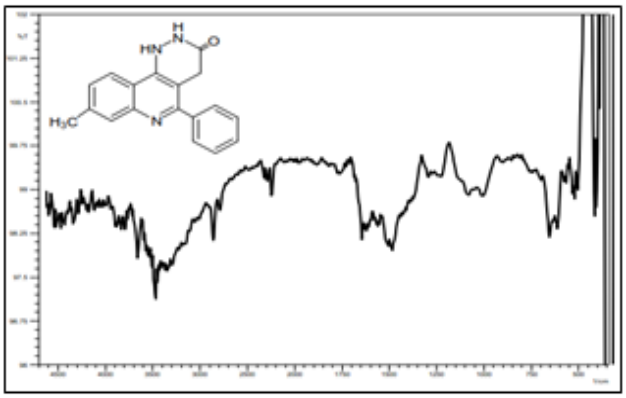


Figure 10: IR spectrum of 8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-c]quinolin-3(4H)-one 7a.

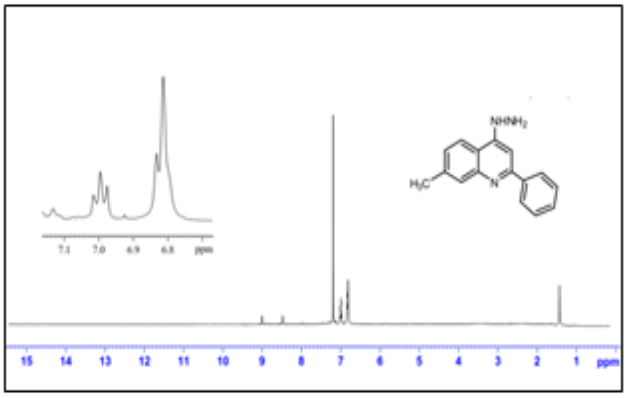


Figure 8: ^{1}H-NMR spectrum of 4-hydrazino- 7-methyl-2-phenyl- quinoline 6a.

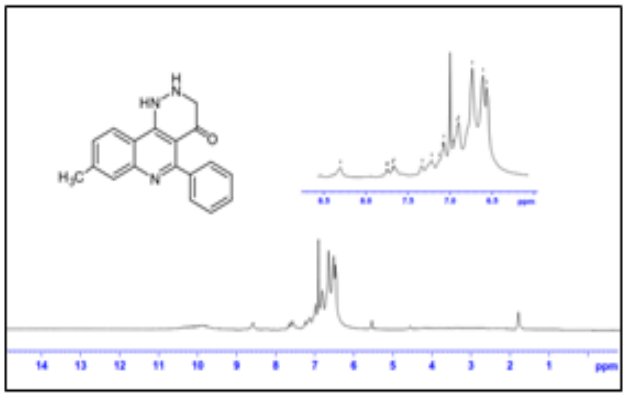


Figure 11: ^{1}H-NMR spectrum of 8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-c]quinolin-3(4H)-one 7a.

Table 1: Antimicrobial activity of substituted pyridazino quinoline derivatives.

	S. aureus				*S. pyogens*			
Conc. in µL	**0.25**	**0.5**	**0.75**	**1**	**0.25**	**0.5**	**0.75**	**1**
	Zone of inhibition diameter in mm							
Compound4a	12	15	17	19	14	16	20	21
Compound4c	12	15	17	20	17	18	19	23
Compound4d	10	11	15	18	18	20	22	24
Compound4e	10	12	16	19	11	13	18	20
Compound4f	11	14	15	17	-	13	18	23
Compound4g	10	10	15	19	12	13	18	22
Compound7a	12	14	15	16	-	19	20	23
Compound7b	11	13	16	18	12	13	18	25
Compound7c	10	14	16	19	16	17	18	20
Compound7d	10	12	16	26	16	20	25	27
Compound7e	10	10	11	12	13	18	20	25

Table 2: Antimicrobial activity of substituted pyridazino quinolones.

	E. coli				*Pseudomonas Sp.*			
Conc. in µL	**0.25**	**0.5**	**0.75**	**1**	**0.25**	**0.5**	**0.75**	**1**
	Zone of inhibition diameter in mm							
Compound4a	13	16	20	23	12	14	15	19
Compound4c	13	15	18	20	11	13	15	17
Compound4d	12	13	20	22	12	15	17	19
Compound4e	10	11	16	18	13	16	18	21
Compound4f	14	16	24	26	13	14	17	21
Compound4g	10	12	13	15	12	14	15	18
Compound7a	10	15	16	17	14	14	16	19
Compound7b	12	15	18	20	12	14	19	26
Compound7c	10	10	15	20	12	14	15	18
Compound7d	10	12	14	16	-	13	17	20
Compound7e	11	11	14	15	15	18	19	21

hydrogen to free radicals to convert it in to an unreactive species. Addition of hydrogen removes the odd electron which is responsible for radical reactivity. The hydrogen-donating activity, of the compounds were measured using DPPH (1,1-diphenyl-2-picrylhydrazyl) radicals scavenging activity. As hydrogen acceptor, showed that a significant association could be found between the concentration of novel molecule and percentage of inhibition.

Antioxidant activity of substituted pyridazino quinolin-3-one was done using DPPH radical scavenging activity method[20] and tabulated.

From Table 3 and Figure 15 9-chloro-5-phenyl-1,2-dihydropyridazino[3,4-*b*]quinolin-3(4*H*)-one 4 f acted as

the best drug as anti-oxidant. The hydrogen donating capacity of the compound is much more than that of the other screened drug as well as the control.

In vivo Studies

In vivo studies are those in which the effect of various biological systems is tested on living organisms or cells usually in animals. Here we have studied the anti-inflammatory and analgesic property of selected compounds which were highly effective in *in vitro* studies.

Anti-inflammatory effect of the 5-phenyl-1,2-dihydropyridazino[4,3-*c*]quinolin-3(4*H*)-one 7d.

To study the effect of drug on inflammation we have chosen the third hour percentage inhibition because the inflammatory effect is maximum at its third hour. At doses of 25 mg/kg body weight 25%, 20.3%, 24.4%, 55%, 56%, and 44.8% inhibition, at 1st 2^{nd} 3^{rd} 4^{th} 5^{th} and 6^{th} hr respectively. As percentage inhibition value at third hour of 7d is comparable with the standard, it may be used as an effective drug towards the inflammations.

Antinociceptive Activity of 5-phenyl-1,2-dihydropyridazino[4,3-*c*]quinolin-3(4*H*)-one 7d

From the Table 4 and Figure 16 it is indicated that the antinociceptive effect of 5-phenyl-1,2-dihydropyridazino[4,3-*c*]quinolin-3(4*H*)-one 7d is 54.88%. The tested compound showed significant reduction in the number of writhes.

Non-linear optical studies

From Table 5,6 and Figure 17 as far as the pyridazinones are concerned linearly fused 8-methyl-5-phenyl-1,2-dihydropyridazino[3,4-*b*]quinolin-3(4*H*)-one 4b exhibited higher value for hyper polarisibility and hence possess a good non-linear optical property as compared to other derivatives.

Among angularly fused substituted pyridazinones, 8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-*c*] quinolin-3(4*H*)-one 7a displayed highest value.

Comparing the hyper polarizability values of linear and angular pyridazinones 8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-*c*]quinolin-3(4*H*)-one 7a was found to be the most effective NLO material.

Instrumental method

Z Scan Technique

When we compare the hyperpolarisability of two sets of pyridazinones, 8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-*c*]quinolin-3(4*H*)-one 7a showed maximum value of hyperpolarisability and thus NLO property. On the basis of above results we performed

Table 3: Antioxidant activity of substituted pyridazino quinolin-3-one.

samples	DPPH Scavenging Activity
Compound 4a	8.75±0.68
Compound 4c	22.41±0.45
Compound 4d	12.14±0.45
Compound 4e	58.14±0.54
Compound 4f	72.54±0.6
Compound 4g	83.45±0.5
Compound 7a	7.35±0.53
Compound 7b	7.16±0.55
Compound 7c	7.19±0.55
Compound 7d	44.74±1.0
Compound 7e	25.14±2.5
Control	26.36±2.5

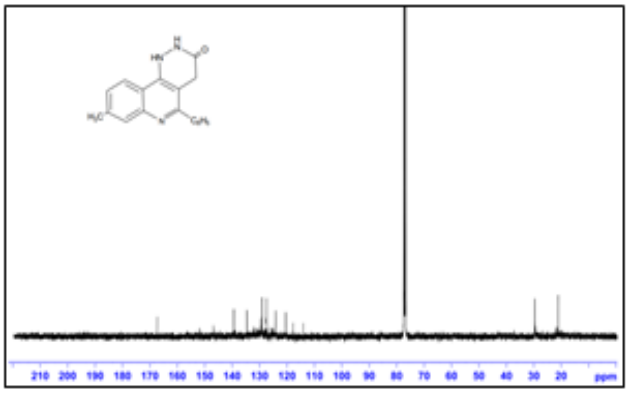


Figure 12: ^{13}C-NMR spectrum of 8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-c]quinolin-3(4H)-one 7a

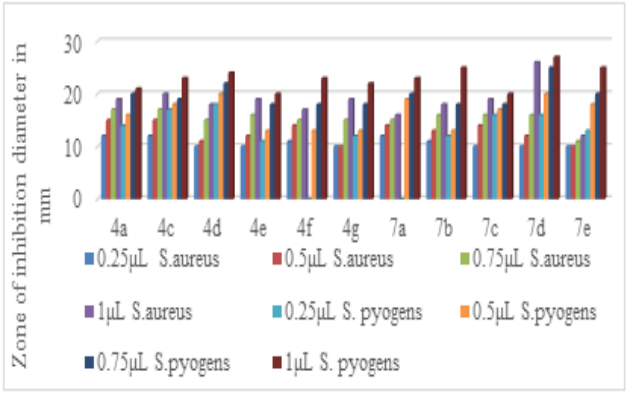


Figure 13: Antimicrobial activity of substituted pyridazino quinoline derivatives.

Table 4: Variation in paw thickness.

	Pre induction	0th hr	1st hr	2nd hr	3rd hr	4th hr	5th hr	6th hr
Control (carrageenan)	1.33±.2	2.48±.2	2.95±.2	3.1±.2	3.37±.4	3.14±.2	2.92±.2	1.96±.1
Drug 7d	1.508±.1	2.568±.2	2.716±.2	2.914±.2	3.044±.3	2.316±.1	2.192±.2	1.85±.2
Std(diclofenac)	1.76±.2	2.79±.2	3.06±.1	3.44±.04	4.09±.3	3.9±.1	3.7±.2	3.57±.1

Table 5: Paw thickness value.

	Variations in paw thickness					
	1st hr	2nd hr	3rd hr	4th hr	5thhr	6thhr
Control (carrageenan)	1.614	1.764	2.032	1.806	1.582	0.62
Drug 7d	1.208	1.406	1.536	0.808	0.684	0.342
Std (diclofenac)	1.554	1.292	1.714	2.025	0.724	0.66

Table 6: % Inhibition value.

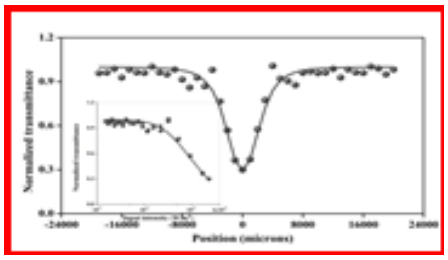
	No. of writhings					Mean	% Inhibition
STD	73	58	48	32	60	54.2 ±15.27	28.49604
Control	80	73	72	87	67	75.8±7.7	
Drug 7d	29	36	38	41	27	54.2 ±15.27	54.88127

the Z scan technique to measure the NLO character of the synthesised compound experimentally.

The Z-scan measurements were done using the standard open aperture Z-scan technique.[17]

In the Z-scan curves the symbols indicate experimental data and the solid lines indicate theoretical fit according to the model for SA associated with two photon absorption.

Measured NLO parameters of 8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-*c*]quinolin-3(4*H*)-one 7a.



β (Non-linear optical coefficient)	2.300X 10^{-10} m/W
I saturation	380.10 X 10^{10} W/m^{-2}
Wavelength	532.00 nm
omega zero	16.50 microns
Pulse width	7000.00 ps
Average Energy	68.89 mjoule
Slices	1000
Linear Transmision	0.43

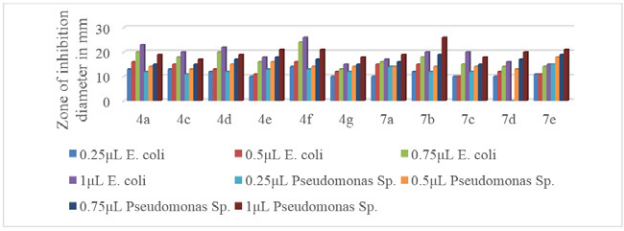


Figure 14: Open aperture Z- scan signatures of 8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-c]quinolin-3(4H)- one 7a.

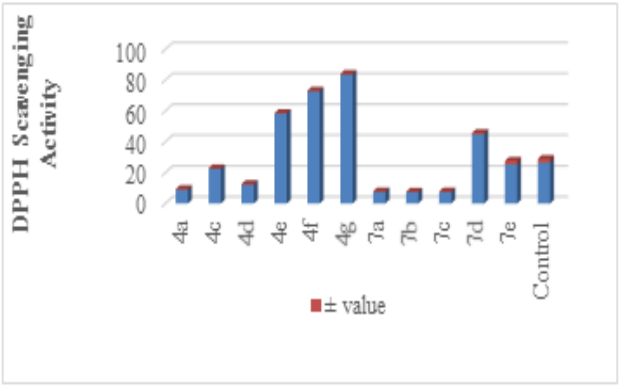


Figure 15: Antioxidant activity of substituted pyridazino quinolin-3-one.

Table 7 Open aperture Z- scan signatures of 8-methyl-5-phenyl-1,2-dihydropyridazino[4,3-c]quinolin-3(4H)-one 7a

The Z- scan parameters obtained for the compound 7a were shown in Table 7 and Figure 14. All the open aperture Z-scan curve was found to fit well with the nonlinear absorption mechanism of 2 photon absorption

Table 7: μ, α and β of Substituted pyridazino quinolones.

Com	μ_{total}	α_{total} esu	β_{total} esu	Com	μ_{total}	α_{total} esu	β_{total} esu
4a	2.27	29.20×10^{-24}	0.50×10^{-30}	7a	7.68	23.25×10^{-24}	2.76×10^{-30}
4b	6.49	28.04×10^{-24}	2.47×10^{-30}	7b	2.99	30.70×10^{-24}	0.73×10^{-30}
4c	5.82	30.28×10^{-24}	1.47×10^{-30}	7c	7.61	21.69×10^{-24}	2.70×10^{-30}
4d	5.14	25.90×10^{-24}	1.83×10^{-30}	7d	2.49	26.46×10^{-24}	0.53×10^{-30}
4e	3.21	36.17×10^{-24}	1.18×10^{-30}	7e	2.17	31.82×10^{-24}	0.87×10^{-30}
4f	2.11	33.34×10^{-24}	0.92×10^{-30}	7f	3.85	30.63×10^{-24}	1.25×10^{-30}
4g	3.44	35.89×10^{-24}	1.13×10^{-30}	7g	3.65	29.48×10^{-24}	1.12×10^{-30}

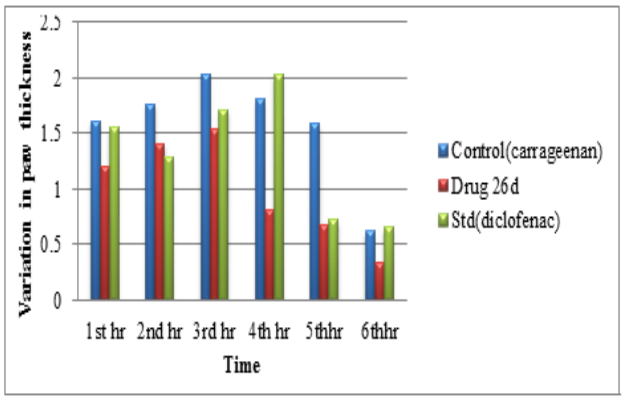


Figure 16: Anti-inflammatory effect.

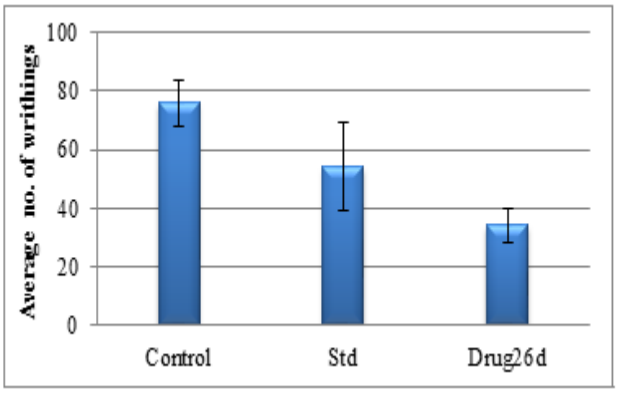


Figure 17: Antinociceptive Effect.

assisted exited state absorption. The observed non-linearity was found to be recognizable.

CONCLUSION

The synthesis and characterization of novel pyridazinones of 4-phenyl and 2-phenyl substituted quinolines were studied and their physical and pharmacological properties were examined. The *in vitro* antibacterial and antioxidant studies were carried out. The results obtained were in good agreement with that of standard. The *in vivo* anti-inflammatory and antinociceptive capability of the selected compound was explained using carrageenan induced acute inflammation model and acetic acid induced writhing model respectively. *In silico* and *Z-scan* instrumental techniques were employed to study the non-linear optical behaviour of the synthesised compounds. *In in silico* method, the second order hyperpolarizibility of the all the compounds were calculated and compared with that of standard urea. Further the *Z scan* technique was utilized to find out the non-linear optical character of the selected synthesised compound experimentally.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

DMSO: Dimethyl Sulphoxide; **TLC:** Thin Layer Chromatography; **IR:** Infra Red; **NMR:** Nuclear Magnetic Resonance; **NLO:** Non-linear Optical; **DMA:** Dimethyl Aniline; **Z:** Zee; **MIC:** Minimum Inhibitory Concentration.

REFERENCES

1. Bourguignon JJ, Oumouch S, Schmitt M. Use of Polyfunctionalized Pyridazines as Reactive Species for Building Chemical Diversity Curr Org Chem. 2006;10:277-95. https://epdf.pub/advances-in-heterocyclic-chemistry-volume-102.html
2. Isabel E. Biological activity and preclinical efficacy of azetidinyl pyridazines as potent systemically-distributed stearoyl-CoA desaturase inhibitors. Bioorg Med Chem Lett. 2011;21:479–83. https://www.sciencedirect.com/science/article/abs/pii/S0960894X10015684
3. Butnariu RM, Mangalagiu II. Mangalagiu, New pyridazine derivatives: synthesis, chemistry and biological activity. Bioorg Med Chem. 2009;17:2823-9. https://europepmc.org/article/med/19272782
4. Gleave RJ, *et al*. Page, Sadhana Patel, Susan Roomans, Brian P. Slingsby, Martin E. Swarbrick, Synthesis and evaluation of 3-amino-6-aryl-pyridazines as selective CB2 agonists for the treatment of inflammatory pain. Bioorg Med Chem Lett. 2010;20:465-8. https://daneshyari.com/article/preview/1375331.pdf

5. Wan Z. Pyridazine-derived γ-secretase modulators. Bioorg Med Chem Lett. 2011;21:4016-9. https://www.sciencedirect.com/science/article/abs/pii/S0960894X11006135
6. Ritchie TJ, Macdonald SJF, Peace S, Pickett SD, Luscombe CN. The developability of heteroaromatic and heteroaliphatic rings – do some have a better pedigree as potential drug molecules than others. Med Chem Commun. 2012;3:1062-9. https://pubs.rsc.org/en/content/articlelanding/2012/md/c2md20111a#!divAbstract
7. Wermuth CG, Schlewer G, Bourguignon JJ, Maghioros G, Bouchet MJ, Moire C, *et al*. 3-Aminopyridazine derivatives with atypical antidepressant, serotonergic and dopaminergic activities. Med Chem. 1989;32:528-37.
8. Volonterio A, Moisan L, Rebek J. Synthesis of Pyridazine-Based Scaffolds as α-Helix Mimetics. Jr Org Lett. 2007;9:3733-6. https://pubs.acs.org/doi/10.1021/ol701487g
9. Isabel E, Powell DA, Black WC, Chan CC, Crane S, Gordon R, *et al*. Biological activity and preclinical efficacy of azetidinyl pyridazines as potent systemically-distributed stearoyl-CoA desaturase inhibitors. Bioorg Med Chem Lett. 2010;21:479-83. http://europepmc.org/article/MED/21074991
10. Wan Z. drianHallYunJinJia-ing Xiang Eric Yang Andrew Eatherton Beverley Smith Guang Yang Haihua Yu Ju Wang Liang Ye Lit-Fui Lau TingYang William Mitchell Wei Cai Xiaomin Zhang Yingxia Sang Yonghui Wang Yasuji Matsuoka, Pyridazine-derived γ-secretase modulators, Bioorg. Med. Chem. Lett. 2011, 21, 4016−4019. https://www.sciencedirect.com/science/article/abs/pii/S0960894X11006135
11. Gleave RJ, *et al*. Synthesis and evaluation of 3-amino-6-aryl-pyridazines as selective CB2 agonists for the treatment of inflammatory pain. Bioorg Med Chem Lett. 2010;20:465-8.
12. Perez C, Pauli M, Bazerque P. Phytochemical Evaluation and in Vitro Antibacterial Activity of Sphaeranthus indicus (L.)-An Important Antijaundice Medicinal Plant. Acta Biologiae et Medecine Experimentalis. 1990;15:113-5. https://www.scirp.org/(S(351jmbntvnsjt1aadkposzje))/reference/ReferencesPapers.aspx?ReferenceID=2017370
13. Blois MS. Antioxidant Determinations by the Use of a Stable Free Radical. Nature. 1958;26:1199-200. https://www.nature.com/articles/1811199a0
14. Liju VB, Jeena K, Kuttan R. An evaluation of antioxidant, anti-inflammatory, and antinociceptive activities of essential oil from *Curcuma longa* L. Ind J Pharmacology. 2011;43(5):526-31. https://www.ijp-online.com/article.asp?issn=0253-7613;year=2011;volume=43; issue=5;spage=526;epage=531;aulast=Liju
15. Gaussian 09W Program, Gaussian Inc., Wallingford, CT. 2009.
16. Adant M, Dupuis M, Bredas JL. Ab initio study of the nonlinear optical properties of urea: Electron correlation and dispersion effects. International Journal of Quantum Chemistry. Int J Quant Chem. 1995;56:497-507. https://onlinelibrary.wiley.com/doi/abs/10.1002/qua.560560853
17. Hausera CND. Georgea. Reynolds. Contribution from the Department of Chemistry, DUKE University. 1948;70.
18. Staskun R, Israelstam SS. Contribution from the Department of Chemistry, University of Witwatersrand. 1961.

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