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Research Article

SYNTHESIS, ANTIMICROBIAL AND ANTIOXIDANT ACTIVITY OF N-DIMETHYLAMINOPHENYLALLYLIDENE CINNAMIDE DERIVATIVES

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ABSTRACT

A series of 2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-(substitutedphenyl)acrylohydrazides**(3a-m)** were synthesized by nucleophilic condensation of various substituted α-benzamido cinnamhydrazides**(2a-m)** with dimethylaminocinnamaldehyde in presence of ethanol and few drops of glacial acetic acid. The chemical structures of synthesized compounds were confirmed by means of IR, ^{1}H NMR, mass spectral and elemental analysis. Among the compounds evaluated, vanillinyl**(3c)** and 4-hydroxy-3,4-dimethoxy derivatives**(3d)** exhibited good antimicrobial activity against all the strains tested, which was comparable to that of standard drugs Ciprofloxacin and Fluconazole and 4-hydroxy-3,4-dimethoxyderivative**(3d)** showed good antioxidant activity towards all the four models. All the derivatives obey Lipinski rule of five and has good bioactive scores.

Keywords: Phenylallylidene, Cinnamides, Antimicrobial activity, Antioxidant activity and *in silico* screening

INTRODUCTION

In recent years, cinnamic acid derivatives such as cinnamides were reported to possess variety of biological activities such as antioxidant[1-2], antimicrobial[3-5], antitumor[6], antitubercular[7-8], anti-inflammatory[9], antifungal[10], anticonvulsant[11] and are often used as promising precursor for the development of new, highly effective drugs. However, the reactive center (vinyl fragment) of cinnamides was significantly affected by substituent present at various positions of the benzene nucleus[12-13]. Hydrazones containing the structure (-CONHN=CH) constitute an important class of compounds for new drug development, which can be synthesized easily in the laboratory by heating appropriate substituted hydrazine or hydrazides with aldehydes / ketones by using various organic solvents. Due to their diverse biological properties like antioxidant[14], anti-inflammatory[15], antibacterial[16], antitumor[17-18], antitubercular[19] etc, hydrazide-hydrazones have gained great importance in the field of medicinal chemistry. The most significant reactivity of the hydrazones is because of nucleophilicity of hydrazone carbon atom.

Bacterial strains are resistant to many currently available antibiotics, so there is urgency for the discovery of new antibacterial agents. Designing of a new lead structure remains a major challenge for researchers. Recently, cinnamide analog bearing hydrazone moieties was reported as significant antibacterial agent[20].

Antioxidants possess the ability to protect the cellular organelles from damage caused by free radical induced oxidation stress. Free radicals include Lipid peroxide, hydroxyl radical, nitric oxide, hydrogen peroxide. Cinnamide analog bearing dimethylaminophenyl moiety has been reported as antioxidant agent[21]. In view of these observations, we have planned to develop a new class of cinnamide derivatives bearing hydrazones and dimethyl amino phenylallylidene moiety with the hope to get potent antimicrobial and antioxidant activities.

MATERIALS AND METHODS

All the melting points reported in this series were determined in open capillaries using Thermonik Precision Melting Point Cum Boiling Point Apparatus C-PMB and are uncorrected. Homogeneity of the compounds was checked by using pre coated Thin Layer Chromatography (TLC) plates. The IR spectra were recorded using KBr pellets technic on FTIR Brucker. ^{1}H-NMR spectra were recorded on Bruker Avance 400 MHz spectrophotometer using Tetramethylsilane (TMS) as an internal standard. Chemical shift (δ) values are reported in δ (ppm). Mass spectra were recorded on an Apex Mass spectrophotometer. All the solvents and chemicals used were procured from Sigma Aldrich, used without further purification.

General method for synthesis of 4-benzylidine-2-phenyloxazol-5-ones (1a-m)

4-Benzylidene-2-phenyloxazol-5-one **(1a)** was prepared by condensing benzaldehyde with benzoyl glycine in presence of acetic anhydride and anhydrous sodium acetate accordance with the previously reported method[22]. The various 4-benzylidene-2-substituted-phenyl-1,3-oxazol-5(4*H*)-ones **(1b-m)** were prepared by similar method.

Synthesis of 2-(benzamido)-3-(aryl)acrylohydrazides (2a-m)

4-Benzylidene-2-phenyloxazol-5(4H)-one **(1a)** (0.03 mol) was stirred with hydrazine hydrate (0.06 mol) in ethanol. The deep yellow colour solid immediately changed to pale yellow colour solid, which was filtered and recrystallised from ethanol accordance with the previously reported method[23]. The various

substituted 2-(benzaido)-3-(aryl)acrylohydrazides **(2b-m)** were prepared by a similar procedure.

Synthesis of 2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-(substitutedphenyl)acrylohydrazide(3a-m)

Equimolar amounts of 2-(benzamido)-3-(aryl)acrylohydrazide and dimethylaminocinnamaldehyde were heated at 60^{o}C with few drops of acetic acid in ethanol for 1 hour. Cool to room temperature, solid formed was filtered and recrytallised from methanol. The various 2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-(substitutedphenyl)acrylohydrazide**(3b-m)** were prepared by similar procedure.

2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-phenylacrylohydrazide (3a)

Yield: 68%; m.p: 170-172°C; IR(KBr)cm^{-}: 3196(N-H), 3029(ArC-H), 1793 & 1655(C=O), 1608(C=C); ^{1}H NMR(400 MHz,DMSO-d$_6$): δ2.95(s,6H,N(CH$_3$)$_2$), 6.70(d,1H,-*CH*=CH-Ar), 6.73(d,1H,-CH=*CH*-Ar), 7.14(s,1H,Ar-CH=C-), 7.32-8.03(m,14H,Ar-H), 8.34(d,1H,N=CH), 10.04(s,1H,NHCO-Ar), 11.42(s,1H,CONHN=); Mass(m/z): 438(M+H)$^{+}$, 437(M-H)$^{-}$; Elemental Analysis: Calculated: C,73.95; H,5.98; N,12.78; O,7.30; Found: C, 73.92; H, 5.96; N, 12.77; O,7.32.

2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-(4-hydroxyphenyl)acrylohydrazide(3b)

Yield: 66%; m.p: 160-161°C; IR(KBr)cm^{-}: 3192(N-H), 3055(ArC-H), 1792&1645(C=O), 1610(C=C); ^{1}H NMR (400 MHz, DMSO-d$_6$): δ 2.85(s,6H,N(CH$_3$)$_2$), 5.75 (s,1H,Ar-OH), 6.74(d,1H,-*CH*=CH-Ar), 6.76(d,1H,-CH=*CH*-Ar),7.10(s,1H,Ar-CH=C-),7.12-7.55(m,13H,Ar-H),8.36(d,1H,N=CH), 10.02(s,1H,NHCO-Ar),11.40(s,1H,CONHN=); Mass (m/z): 454(M+H)$^{+}$, 453(M-H)$^{-}$; Elemental Analysis: Calculated: C,71.35; H,5.77; N,12.33; Found: C,71.38; H, 5.75; N, 12.32.

2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-(4-hydroxy-3-methoxyphenyl)acrylohydrazide(3c)

Yield: 61%; m.p: 172-174°C; IR(KBr)cm^{-}: 3188(N-H), 3054(ArC-H), 1796&1656(C=O), 1602(C=C); ^{1}H NMR (400 MHz, DMSO-d$_6$): δ 2.65(s,6H,N(CH$_3$)$_2$), 3.32(s,3H,OCH$_3$), 5.82(s,1H,Ar-OH), 6.70(d,1H,-*CH*=CH-Ar),6.73(d,1H,-CH=*CH*-Ar),7.11(s,1H,Ar-CH=C-,)6.86-7.66(m,12H,Ar-H), 8.38(d,1H,N=CH), 10.08(s,1H,NHCO-Ar), 11.42(s,1H,CONHN=); Mass(m/z): 484(M+H)$^{+}$, 483(M-H)$^{-}$; Elemental Analysis:Calculated: C, 69.41; H, 5.82; N, 11.56Found: C, 69.42; H, 5.80; N, 11.54.

2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-(4-hydroxy-3,5-dimethoxyphenyl)acrylohydrazide(3d)

Yield: 60%; m.p: 184-185°C; IR(KBr)cm^{-}: 3199(N-H), 3058(ArC-H), 1794&1645(C=O), 1600(C=C); ^{1}H NMR (400 MHz, DMSO-d$_6$): δ 2.50(s,6H, N(CH$_3$)$_2$), 3.80(s,6H,(OCH$_3$)$_2$), 5.78(s,1H,Ar-OH), 6.72(d,1H,-*CH*=CH-Ar), 6.74(d,1H,-CH=*CH*-Ar), 6.85-7.62(m,11H,Ar-H), 7.13(s,1H,Ar-CH=C-),8.36(d,1H,N=CH), 10.14(s,1H,NHCO-Ar), 11.44(s,1H,CONHN=); Mass (m/z): 514(M+H)$^{+}$, 513(M-H)$^{-}$; Elemental Analysis: Calculated: C, 67.69; H, 5.88; N, 10.89 Found: C, 67.70; H, 5.86; N, 10.88.

2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-(4-methoxyphenyl)acrylohydrazide(3e)

Yield: 65%; m.p: 135-136°C; IR(KBr)cm^{-}: 3234(N-H), 3050(ArC-H), 1788&1641(C=O), 1603(C=C); ^{1}H NMR (400 MHz, DMSO-d$_6$): δ 2.95(s,6H,N(CH$_3$)$_2$), 3.76(s,3H,OCH$_3$), 6.70(d,1H,-*CH*=CH-Ar), 6.72(d,1H,-CH=*CH*-Ar), 6.78-7.15(m,13H,Ar-H), 7.15(s,1H,Ar-CH=C-), 8.12(d,1H,N=CH), 9.96(s,1H,NHCO-Ar), 11.34(s,1H,CONHN=); Mass (m/z): 468(M+H)$^{+}$, 467((M-H)$^{-}$; Elemental Analysis: Calculated: C, 71.78; H, 6.02; N, 11.96 Found: C, 71.76; H, 6.04; N, 11.74.

2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-(3,4-dimethoxyphenyl)acrylohydrazide(3f)

Yield: 63%; m.p: 150-152°C; IR(KBr)cm^{-}: 3194(N-H), 3055(ArC-H), 1794&1654(C=O), 1609(C=C); ^{1}H NMR (400 MHz, DMSO-d$_6$):δ 2.68(s,6H,N(CH$_3$)$_2$), 3.51(s,6H,(OCH$_3$)$_2$), 6.72(d,1H,-*CH*=CH-Ar), 6.75(d,1H,-CH=*CH*-Ar),7.10(s,1H,Ar-CH=C-), 7.13-7.50(m,12H,Ar-H),8.36(d,1H,N=CH), 10.08(s,1H,NHCO-Ar),11.48(s,1H,CONHN=); Mass(m/z): 498(M+H)$^{+}$,497(M-H)$^{-}$; Elemental Analysis: Calculated: C, 69.86; H, 6.06; N, 11.24 Found: C, 69.84.54; H, 6.04; N, 11.26.

2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-(3,4,5- trimethoxyphenyl)acrylohydrazide(3g)

Yield: 59%; m.p:165-167°C; IR(KBr)cm^{-}: 3238(N-H), 3060(ArC-H), 1790&1656(C=O), 1601(C=C); ^{1}H NMR (400 MHz,DMSO-d$_6$): δ2.45(s,6H,N(CH$_3$)$_2$), 3.81(s,9H,(OCH$_3$)$_3$), 6.74(d,1H,-*CH*=CH-Ar), 6.76(d,1H,-CH=*CH*-Ar), 6.95(s,1H,Ar-CH=C-), 6.90-7.20(m,11H,Ar-H), 8.38(d,1H,N=CH), 10.10(s,1H,NHCO-Ar), 11.40(s,1H,CONHN=); Mass (m/z): 528(M+H)$^{+}$, 527(M-H)$^{-}$; Elemental Analysis: Calculated: C, 68.17; H, 6.10; N, 10.60 Found: C, 68.18; H, 6.12; N, 10.62.

2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-(4-methylphenyl)acrylohydrazide(3h)

Yield: 67%; m.p: 254-256°C; IR(KBr)cm^{-}:3186(N-H), 3054(ArC-H), 1785&1660(C=O), 1605(C=C); ^{1}H NMR (400 MHz,DMSO-d$_6$): δ2.33(s,3H,CH$_3$), 2.69(s,6H,N(CH$_3$)$_2$), 6.72(d,1H,-*CH*=CH-Ar), 6.74(d,1H, -CH=*CH*-Ar), 6.92(s,1H,Ar-CH=C-), 7.18-7.42(m,13H,Ar-H), 8.24(d,1H,N=CH), 10.01(s,1H,NHCO-Ar), 11.48(s,1H, CONHN=); Mass (m/z): 452(M+H)$^{+}$, 451(M-H)$^{-}$; Elemental Analysis: Calculated: C, 74.31; H, 6.24; N, 12.38 Found: C, 74.30; H, 6.22; N, 12.36.

2-(benzamido)-3-(4-(dimethylamino)phenyl)-N-(3-(4-dimethylamino)phenyl)allylidene) acrylohydrazide(3i)

Yield: 69%; m.p: 245-247°C; IR(KBr)cm^{-}: 3395(N-H), 3021(ArC-H), 1760&1651(C=O), 1602(C=C); ^{1}H NMR(400 MHz, DMSO-d$_6$): δ2.50(s,6H,N(CH$_3$)$_2$), 2.93(s,6H,N(CH$_3$)$_2$), 6.67(d,1H,-*CH*=CH-Ar), 6.94(s,1H,Ar-CH=C-), 6.98-7.61(m,13H,Ar-H), 8.06-8.08(d,1H,-CH=*CH*-Ar), 8.18(d,1H,N=CH), 10.03(s,1H,NHCO-Ar), 11.48(s,1H,CONHN=); Mass (m/z): 481(M+H)$^{+}$, 480(M-H)$^{-}$; Elemental Analysis: Calculated: C,72.33; H, 6.49; N,14.54 Found: C, 72.34; H, 6.48; N, 14.53.

2-(benzamido)-3-(4-chlorophenyl)-N-(3-(4-dimethylamino)phenyl)allylidene) acrylohydrazide(3j)

Yield: 58%; m.p: 180-182°C; IR(KBr)cm^{-}: 3246(N-H), 3060(ArC-H), 1780&1645(C=O), 1606(C=C); ^{1}H NMR (400 MHz, DMSO-d$_6$): δ 2.45(s,6H,N(CH$_3$)$_2$), 6.72(d,1H,-*CH*=CH-Ar), 6.73(d,1H,-CH=*CH*-Ar), 6.90(s,1H,Ar-CH=C-), 7.12-7.80(m,13H,Ar-CH), 8.40(d,1H,N=CH), 10.08(s,1H,NHCO-Ar), 11.48(s,1H, CONHN=); Mass (m/z): 472(M+H)$^{+}$, 471(M-H)$^{-}$; Elemental Analysis: Calculated: C, 68.56; H, 5.33; N, 11.85 Found: C, 68.54; H, 5.31; N, 11.86.

2-(benzamido)-3-(4-cyanophenyl)-N-(3-(4-dimethylamino)phenyl)allylidene)acrylohydrazide(3k) Yield: 62%; m.p: 178-179°C; IR(KBr)cm^{-}: 3185(N-H), 3056(ArC-H), 1785&1665(C=O), 1612(C=C); ^{1}H NMR (400 MHz, DMSO-d$_6$): δ 2.62(s,6H,N(CH$_3$)$_2$), 6.75-6.86(d,1H,-*CH*=CH-Ar), 6.73(d,1H,-CH=*CH*-Ar), 7.12(s,1H,Ar-CH=C-), 7.10-7.84(m,13H,Ar-CH), 8.36(d,1H,N=CH), 10.10(s,1H,NHCO-Ar), 11.45(s,1H, CONHN=); Mass (m/z): 463(M+H)$^{+}$, 462(M-H)$^{-}$; Elemental Analysis: Calculated: C, 72.55; H, 5.44; N, 15.11 Found: C, 72.53; H, 5.42; N, 15.10.

2-(benzamido)-N-(3-(4-dimethylamino)phenyl)allylidene)-3-(4-nitrophenyl)acrylohydrazide(3l)

Yield: 65%; m.p: 175-177°C; IR(KBr)cm^{-}: 3228(N-H), 3058(ArC-H), 1790&1652(C=O), 1601(C=C); ^{1}H NMR (400 MHz, DMSO-d$_6$): δ 2.70(s,6H,N(CH$_3$)$_2$), 6.68(d,1H,-*CH*=CH-Ar), 6.73(d,1H,-CH=*CH*-Ar), 7.10(s,1H,Ar-CH=C-), 7.14-7.92(m,13H,Ar-H), 8.35(d,1H,N=CH), 10.24(s,1H,NHCO-Ar), 11.42(s,1H, CONHN=); Mass (m/z): 483(M+H)$^{+}$, 482(M-H)$^{-}$; Elemental Analysis: Calculated: C, 67.07; H, 5.21; N, 14.48 Found: C, 67.09; H, 5.20; N, 14.47.

2-(benzamido)-N-(3-(4-dimethylamino)phenyl)allylidene)-3-(3-nitrophenyl)acrylohydrazide(3m) Yield: 68%; m.p:164-166°C; IR(KBr)cm^{-}: 3253(N-H), 3055(ArC-H), 1704&1661(C=O), 1618(C=C);^{1}H NMR (400 MHz, DMSO-d$_6$):δ 2.95(s,6H,N(CH$_3$)$_2$), 6.72(d,1H,-*CH*=CH-Ar), 6.74(d,1H,-CH=*CH*-Ar), 7.11(s,1H,Ar-CH=C-), 7.15-7.94(m,13H,Ar-H), 8.37(d,1H,N=CH), 10.25(s,1H,NHCO-Ar), 11.28(s,1H, CONHN=); Mass (m/z): 483(M+H)$^{+}$, 482(M-H)$^{-}$; Elemental Analysis: Calculated:C, 67.07; H,5.21; N, 14.48 Found: C, 67.08; H, 5.20; N,14.50.

ANTIMICROBIAL ACTIVITY

All the test compounds**(3a-m)** were screened for antimicrobial and antifungal activities by using cultures of gram positive bacteria: *Staphylococcus aureus* , Gram negative bacteria: *Escherichia coli* and fungal strains *Penicillium chrysogenum, Penicillium notatum* and *Aspergillus niger*. The antimicrobial activity was assayed biologically using agar well diffusion method[24]**.** In this method, wells of standard diameter are made in the nutrient agar medium, containing standard bacterial inoculum. The test compounds were introduced into the wells and diameter of the zone of inhibition was measured by antibiotic zone reader. The standard drugs used were ciprofloxacin and fluconazole.

ANTIOXIDANT STUDY

Inhibition of Lipid Peroxidation Method

The brain tissue of fasted rat was weighed and homogenate (10% w/v) was prepared in 0.15 M KCl and centrifuged at 800 rpm for 10 min using homogenizer. The supernatant liquid was used immediately for the study and used as a source of polyunsaturated fatty acids. The extent of lipid peroxidation in rat brain homogenate was measured invitro in terms of formation of thiobarbituric acid reactive substances (TBARS). 1 ml of 100 µM of test solutions was individually added to 0.5 ml of brain homogenate and this mixture was incubated with 0.15 M KCl (100 µl). Lipid peroxidation was initiated by adding 100 µl of 15 mM FeSO4 solution and the reaction mixture was incubated at 37oC for 30 min. An equal volume of TBA: TCA (1:1), 1 ml was added to the above solution followed by the addition of 1 ml BHT. This final mixture was heated on a water bath for 20 min at 80oC, cooled, centrifuged and absorbance was read at 532 nm using a spectrophotometer and all the tests were carried out in triplicate[25].

Determination of the effect of samples on 1, 1-diphenyl-2-picrylhydrazyl (DPPH) Radical

The hydrogen atom or electron donation capability (antioxidant capacity) of the compounds was measured from the bleaching of alcoholic purple colored solution of 1, 1diphenyl-1-picrylhydrazyl (DPPH) and is the valid method to estimate the antioxidant capacity of phenolic compounds. 2ml of standard drug, Ascorbic acid and title compounds (100 µM) were added to 2 ml of 100 µM alcoholic DPPH solution. The tubes were kept at an ambient temperature for 20-30 min protected from light by covering with aluminum foil and absorbance was measured at 517 nm using ethanol as blank. The percentage of inhibition of free radical production from DPPH was calculated by the following equation and all the tests were carried out in triplicate[26].

$$\% \text{ of scavenging} = [(\text{control} - \text{sample}) / \text{control}] \times 100 \rightarrow \textbf{(1)}$$

Where control is the absorbance of the control reaction (containing all reagents except the test compound) and sample is the absorbance of the test compound.

Determination of nitric oxide Scavenging Activity

Nitric oxide scavenging activity of samples was determined by the following procedure. 2ml (10mM) of sodium nitro prusside dissolved in 1.5ml phosphate buffer saline **(PH-7.4)** and 1 ml of different test samples corresponding to 100µM concentration was added in different test tubes respectively and incubated at 25°C; for about 150 min. From this 0.5ml was taken and 1ml sulphanilic acid reagent (33% in 20% glacial acetic acid) was added and incubated at room temperature for 5min. 1ml of naphthyl ethylene diamine dihydro chloride **(0.1% w/v)** was added and again incubated at room temperature for 30min, then measured the absorbance at 540 nm in spectrophotometer[27]. Nitric oxide scavenging activity was calculated by using Eq. **(1).**

Determination of Hydrogen Peroxide Scavenging Activity

4mM solution of H_2O_2 was prepared in phosphate-buffered saline **(PBS, pH 7.4).** H_2O_2 concentration was determined spectrophotometrically from absorbance at 230 nm using molar absorptivity 81 M^{-1} cm^{-1}.1 ml of different samples corresponding to 100µM concentration were added to 0.6ml hydrogen peroxide-PBS solution respectively and control without sample. Absorbance of H_2O_2 at 230nm was determined 10 minutes later against a blank solution. The percentage of scavenging of H_2O_2 was calculated by using Eq. **(1)**[28].

MOLECULAR DESCRIPTORS AND DRUGLIKENESS

In-silico ADME

In the present study molecular properties of compounds **(3a-m)** were calculated by using Molinspiration online tool[29] and SwissADME[30] in order to predict the compounds drug likeness score. The %ABS was calculated according to the formula

$$\%ABS = 109-(0.345 \times TPSA).$$

Some physicochemical parameters of the synthesized compounds were predicted and listed in Table **(3&4).** The predicted values revealed that the compounds obeyed Lipinski rule of five by possessing not more than 5 hydrogen bond donors (OH and NH groups), not more than 10 hydrogen bond acceptors (notably N and O) and not more than 15 rotatable bonds (rotb), a partition coefficient log P was found to be not more than 5 for some compounds. However, descriptors such as log P and solvent accessible surface area contributed towards the activity of the molecules, TPSA is another key property that has been linked to bioavailability. It was found that passively absorbed molecules with a TPSA more than 140 are thought to have low oral availability. TPSA obtained for the tested compounds were below 140 predict good oral bioavailability. Drug likeness scores of the synthesized compounds were predicted using molsoft website. This result is adding support to the biological activities observed for these compounds. Ability to predict the percent oral absorption was the primary goal in the design, optimization, and selection of potential candidates in the development of oral drugs.

BIOACTIVITY SCORE PREDICTION

The bioactivity scores of the compounds **(3a-m)** were calculated for their GPCR ligand, kinase inhibitor, protease inhibitor and enzyme inhibitor activities **(Table 5).** For average organic molecules the probability is that if the bioactivity score is more than 0 then it is active, if 0.5 to 0 then moderately active. Kinase inhibitor and enzyme inhibition scores of -0.52 and -0.29 obtained for compound **4** tested supported the presence of moderate anti-inflammatory and analgesic activity. The protease inhibitor score of compounds **5** was predicted as -0.37 and for compound **4**, -0.27 that is comparable with score of standard protease inhibitor, ritonavir (0.47). Good protease inhibitor scores obtained predicted the efficiency of these compounds as good antimicrobials and might possess protease inhibitor activities.

RESULTS AND DISCUSSION

Chemistry

Thirteen new compounds 2-(benzamido)-N-(3-(4-(dimethylamino)phenyl)allylidene)-3-(substitutedphenyl)acrylohydrazide**(3a-m)** were synthesized and the reaction sequence for the synthesis is outlined in Scheme. The intermediate 2-(benzamido)-3-(aryl)acrylohydrazides **(2a-m)** upon nucleophilic addition with dimethylaminocinnamaldehyde in ethanol and a few drops of acetic acid yielded the title compounds **(3a-m) .** The intermediates **(2a-m)** was obtained by stirring 4-benzylidene-2-phenyloxazol- 5 -one **(1a-m)** with hydrazine hydrate (99%) at room temperature in the presence of ethanol. The compounds **(1a-m)** were synthesized by condensation of Benzoyl glycine with different aromatic aldehydes in accordance with the previously reported method. Spectral data of all the newly synthesized compounds were in full agreement with the proposed structures. The IR spectral data of titled compounds**(3a-m)** displaced bands in the region of 3185-3395cm^{-1} due to N-H stretching, 3025-3060cm^{-1} due to the aromatic C-H stretching, 1649-1800cm^{-1} due to C=O stretching and bands at 1600-1650cm-1 due to C=C stretching. The ^{1}H NMR spectra of the compounds showed multiplet at δ 6.85-7.95 due to aryl protons, doublet at δ 6.90-7.15 due to styryl protons and 6.72-8.40 due to cinnamyl protons, singlet at δ10.04 due to NHCO and singlet at δ 11.42 due to – CONHN=. The mass spectrum of compounds **3a** and **3m** showed the characteristic molecular ion peak(M±H) at m/z 438and483 respectively. The Elemental analyses of the compounds were found to be within the limits of ±0.4% of theoretical values.

Anti-microbial activity

The anti-microbial data of title compounds was tabulated (Table 1). Among the series, 4-hydroxy-3-methoxy**(3c)** and 4-hydroxy-3,5-dimethoxy**(3d)** derivatives exhibited good anti-microbial activity towards all the organisms.These results are in agreement with earlier reports indicating that phenolic compounds containing methoxy substituents in ortho position showed good amtimicrobial activity[31].The unsubstituted**(3a)**, 4-hydroxy**(3b)**, 4-methoxy**(3e)**, 3,4-dimethoxy**(3f)**, 3,4,5-trimethoxy**(3g)**, 4-N,Ndimethyl**(3i),** 4-cyano**(3k)** and 4-nitro**(3l)** derivatives exhibited moderate antimicrobial activity towards all the organisms.The 4-methyl**(3h)** and 4-chloro**(3j)** derivatives was found to be moderatly active towards *E.coli*. The 4-chloro**(3j)** and 3-nitro**(3m)** derivatives showed moderate activity towards *P.notatum* and *A.niger*.

Antioxidant activity

Inhibition of Lipid Peroxidation Method

All the compounds **(3a-m)** were screened by lipid peroxidation method. Among the series obtained,compounds **3d (65%)** and **3i (63%)** exhibited good antioxidant activity when compared to the standard tocopherol **(61%).** Compounds **3k (60%)** was found to be equipotent to the standard. From **(Table 2)** we came to know the sequence of the inhibition of lipid peroxidation was found to be 3g > 3f > 3e indicates the contribution of EDG (alkoxy) groups towards the antioxidant activity i.e. increasing the number of methoxy groups **(3g (44%))** enhances the antioxidant activity. On the basis of structure activity relationship, the presence of EDG on electron rich aryl systems **(3c, 3g, 3h, 3f and 3e)** essential for the antioxidant activity and was also observed when compared to EWG **(3l)** as substitution.

DPPH Scavenging Method

The free radical scavenging capacity of the compounds **(3a-m)** depends upon a hydrogen atom transfer (HAT) to DPPH free radical. DPPH is a stable free radical, it shown strong absorption band at 515 nm due to its odd electron configuration and decreases the absorbance in presence of free radical scavengers resulting in a color change from deep purple to yellow[33-34]. All the compounds exhibited radical scavenging activity. Among those compounds **3d (58%)** and **3c (55%)** showed good antioxidant activity. Compounds **3i (54%)** and **3k (51%)** elicited comparable activity to that of standard.

Nitric Oxide Method

The results from **(Table 2)** revealed that compounds **3d,** and **3c** showed good antioxidant activity compared with the standard. This might be due to the presence of electron donating groups .

Hydrogen Peroxide Method

All the compounds exerted moderate to good activity against hydrogen peroxide method. Among the compounds **3d (54%)** and **3i (48%)** showed good antioxidant activity.

***In silico* ADME**

All the compounds obeyed Lipinski rule of five is important for assessing of compounds oral bioavailability. It is clear from the **(Table 3)** that log P values of all the compounds found to be in the acceptable criteria (3.96-5.90) and TPSA (Total polar surface area) (<140) is another key property that has been linked to bioavailability. It was found that all the compounds showed good TPSA predicts good oral bioavailability. The result of in silico data indicates that, these compounds may have the potential to become a lead compound.

Table 1: Antimicrobial activity of title compounds (3a-m)

		Diameter of zone of inhibition(mm)				
Compound Code	**Compound**	***E.coli***	***S.aureus***	***P.notatum***	***P.chrysogenum***	***A.niger***
3a	C_6H_5	12	12	8	10	10
3b	4-OH C_6H_4	16	16	12	NA	NA
3c	4-OH ,3-OCH_3 C_6H_3	18	20	20	20	20
3d	4-OH ,3,5 -(OCH_3)$_2$ C_6H_2	18	18	20	16	16
3e	4- OCH_3 C_6H_4	18	19	10	7	7
3f	3,4 –(OCH_3)$_2$ C_6H_3	12	16	16	19	10
3g	3,4,5 -(OCH_3)$_3$ C_6H_2	18	16	12	14	12
3h	4- CH_3	10	NA	NA	NA	NA
3i	4-N,N(CH_3)$_2$ C_6H_4	17	16	14	16	12
3j	4-Cl C_6H_4	9	NA	9	NA	10
3k	4-CN C_6H_4	12	12	10	10	7
3l	4-NO_2 C_6H_4	11	12	10	10	18
3m	3-NO_2 C_6H_4	10	10	17	11	17
Fluconazole		-	-	23	23	21
Ciprofloxacin		20	19	-	-	-

NA-Not Active**,** a. Concentration of 100 µg/ml for Test compounds and standards (ciprofloxacin and Fluconazole respectively), b. Control -DMSO, c. Activity is measured as zone of inhibition in mm

Table 2: Antioxidant activity for title compounds (3a-m)

Compound Code	**Compound**	**LP %Inhibition at 100µM**	**DPPH %Inhibition at 100µM**	**NO %Inhibition at 100µM**	**H_2O_2 %Inhibition at 100µM**
3a	C_6H_5	33±0.6	36±0.5	45±0.8	29±0.6
3b	4-OH C_6H_4	32±0.6	31±1.5	35±0.5	21±1.5
3c	4-OH ,3-OCH_3 C_6H_3	47±1.3	55±0.9	68±0.8	41±1.1
3d	4-OH ,3,5 -(OCH_3)$_2$ C_6H_2	65±0.6	58±0.6	70±1.0	54±0.4
3e	4- OCH_3 C_6H_4	27±0.9	25±1.7	21±1.3	19±1.5
3f	3,4 –(OCH_3)$_2$ C_6H_3	33±0.7	33±0.8	40±0.5	22±1.5
3g	3,4,5 -(OCH_3)$_3$ C_6H_2	44±0.6	43±0.9	47±1.0	28±1.6
3h	4- CH_3	41±0.4	40±1.1	42±0.8	28±1.3
3i	4-N,N(CH_3)$_2$ C_6H_4	63±0.5	54±0.8	51±0.9	48±0.9
3j	4-Cl C_6H_4	32±0.6	20±0.8	20±1.6	18±0.4
3k	4-CN C_6H_4	60±3.5	51±0.6	50±1.0	44±1.0
3l	4-NO_2 C_6H_4	30±1.0	27±0.9	24±0.4	20±0.9
3m	3-NO_2 C_6H_4	36±1.3	37±1.4	36±0.4	21±1.9
Tocopherol		61±0.9	52±0.7	-	-
Ascorbic acid		-	-	69±0.6	49±0.8

- : Not tested; LP: Lipid peroxidation; DPPH: Diphenyl picryl hydrazine; NO: Nitric oxide; H_2O_2: Hydrogen peroxide methods; Results are presented as Mean±SEM in Triplicate.

Table 3: Molecular property prediction of the title compounds (3a-m) using molinspiration.com

Compound Code	**Compound**	**MW[a]**	**LogP[b]**	**TSPA[c]**	**noN[d]**	**noHNH[e]**	**Nrotb[f]**	**Vol[g]**	**nAtoms[h]**
3a	C_6H_5	438.53	4.99	73.80	6	2	8	413.63	63
3b	4-OH C_6H_4	454.53	4.51	94.03	7	3	8	421.65	34
3c	4-OH ,3-OCH_3 C_6H_3	484.56	4.33	103.26	8	3	9	447.20	36
3d	4-OH ,3,5 -(OCH_3)$_2$ C_6H_2	514.58	4.34	112.49	9	3	10	472.74	38
3e	4- OCH_3 C_6H_4	468.58	5.05	83.03	7	2	9	439.10	35
3f	3,4 –(OCH_3)$_2$ C_6H_3	498.58	4.64	92.27	8	2	10	464.70	37
3g	3,4,5 -(OCH_3)$_3$ C_6H_2	528.61	4.62	101.5	9	2	11	490.27	39
3h	4- CH_3	452.56	5.44	73.80	6	2	8	430.19	34
3i	4-N,N(CH_3)$_2$ C_6H_4	481.60	5.09	77.04	7	2	9	459.54	34
3j	4-Cl C_6H_4	472.98	5.67	73.80	6	2	8	427.17	34
3k	4-CN C_6H_4	463.54	4.75	97.59	7	2	8	430.49	35
3l	4-NO_2 C_6H_4	483.53	4.95	119.62	9	2	9	436.97	36
3m	3-NO_2 C_6H_4	483.53	4.92	119.62	9	2	9	436.97	36

a.Molecular weight, b.Log P – Partition coefficient, c.TPSA-Topological polar surface area, d.NON- No. of hydrogen bond acceptors, e.NOHNH- No. of hydrogen bond donors, f.Nrotb- No. of rotatable bonds, g.Volume, h.N atoms.

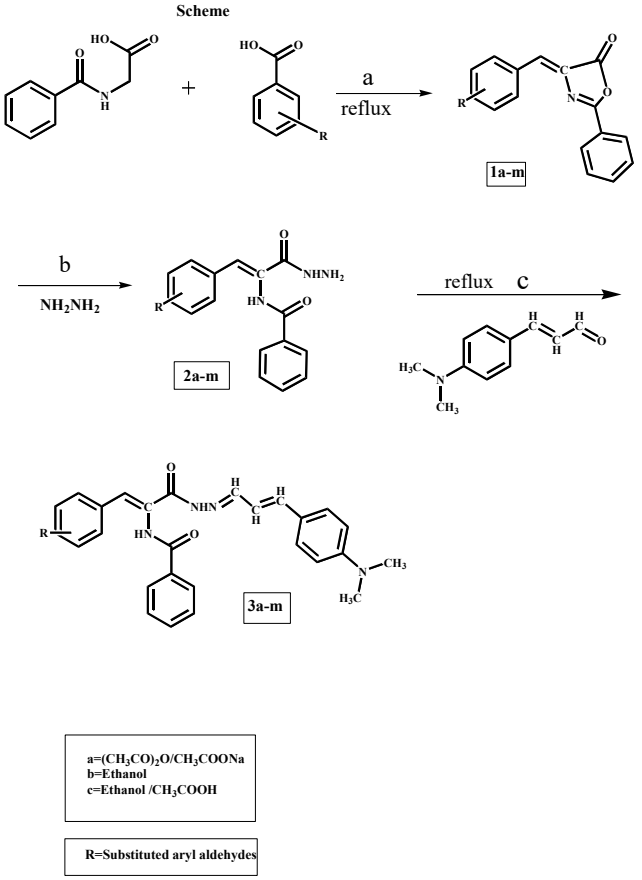
Table 4: Molecular property prediction of the title compounds (3a-m) using SWISSADME

Compound code	Lipinski rule of five	BBB	Bioavailability	Ghose filter	Muegge filter	Veber filter
3a	YES	YES	0.55	NO	YES	YES
3b	YES	NO	0.55	NO	YES	YES
3c	YES	NO	0.55	NO	YES	NO
3d	YES	NO	0.55	NO	YES	NO
3e	YES	NO	0.55	NO	YES	NO
3f	YES	NO	0.55	NO	YES	NO
3g	YES	NO	0.55	NO	YES	NO
3h	YES	YES	0.55	NO	NO	YES
3i	YES	NO	0.55	NO	YES	NO
3j	YES	NO	0.55	NO	NO	YES
3k	YES	NO	0.55	NO	YES	YES
3l	YES	NO	0.55	NO	YES	NO
3m	YES	NO	0.55	NO	YES	NO

Table 5: Bioactivity scores of title compounds (3a-m)

Compound code	GPCR Ligand	Ion Channel Modulator	Kinase Inhibitor	Nuclear Receptor Ligand	Protease Inhibiter	Enzyme Inhibiter
3a	-0.32	-0.69	-0.57	-0.75	-0.37	-0.31
3b	-0.28	-0.64	-0.52	-0.63	-0.35	-0.27
3c	-0.31	-0.70	-0.52	-0.67	-0.40	-0.29
3d	-0.31	-0.74	-0.50	-0.66	-0.37	-0.27
3e	-0.34	-0.71	-0.56	-0.72	-0.39	-0.33
3f	-0.33	-0.74	-0.53	-0.70	-0.40	-0.32
3g	-0.32	-0.81	-0.53	-0.74	-0.39	-0.34
3h	-0.34	-0.72	-0.58	-0.75	-0.40	-0.35
3i	-0.29	-0.67	-0.52	-0.69	-0.34	-0.29
3j	-0.32	-0.68	-0.57	-0.75	0.39	-0.33
3k	-0.29	-0.66	-0.46	-0.64	-0.34	-0.27
3l	-0.40	-0.70	-0.63	-0.75	-0.44	-0.36
3m	-0.41	-0.71	-0.62	-0.76	-0.44	-0.37

Scheme



CONCLUSION

In the present study a new series of Cinnamide derivatives were synthesized and screened for antimicrobial and antioxidant activity. Among the series, compound **(3c)** and **(3d)** exhibited good antibacterial, antifungal activities and compound **(3d)** showed antioxidant activity. From the results it was evident that the further modification of the substituents on the benzylidene moiety gives a new lead molecule.

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REFERENCES

1. Eleni P, Dimitra HL, Konstantinos L, George G. Novel cinnamic acid derivatives as antioxidant and anticancer agents: Design, synthesis and modeling studies. Molecules. 2014; 19: 9655-9674.
2. Gangadhara S, Prasad CH, Venkateswarlu P. Synthesis, antimicrobial and antioxidant activities of novel series of cinnamamide derivatives having morpholine moiety. Medicinal chemistry. 2014; 4(12): 778-783.
3. Juan David G. Natural cinnamic acids: Synthetic derivatives and hybrids with antimicrobial activity. Molecules. 2014; 19: 19292-19349.
4. Xiaoda S, Yushun Y, Jing Z, Yangjian C. Synthesis and anti-bacterial activity of Cinnamaldehyde Acylhydrazone with a 1,4-Benzodioxan fragment as a Novel Class of potent β-Ketoacyl-Acyl Carrier Protein synthase III (FabH) inhibitor. Chemical and Pharmaceutical Bulletin.2014; 62(11): 1110-1118.
5. Saritha K, Rajitha G, SudheerKumar K, Umamaheswari A. Synthesis, Molecular Docking and Antimicrobial activity of substituted Cinnamides. International journal of Pharmacy and Biological Sciences. 2018; 8(3): 770-778.
6. Hadjipavlou-Litina D and Pontiki E. Aryl acetic and cinnamic acids as lipoxygenase inhibitors with antioxidant, anti-inflammatory and anticancer activity. Methods in Molecular Biology. 2015; 1208: 361-377.
7. Samir A, Edson F da silva, Marcus V N, desouza Maria C S Laurenco, Felipe R V. Synthesis and antimycobacterial evaluation of new trans-Cinnamic acid hydrazide derivatives. Bioorganic and Medicinal Chemistry Letters. 2008; 18: 538-541.
8. Manoj D Kakwani, Prashant Surya vamsi, Mukti kant Ray, M G Rajan, Sharmila Majee, Abdul Samad, Padma Devarajan, MariamS.Degani. Design,synthesis and antimycobacterial activity of cinnamide derivatives. Bioorganic and Medicinal Chemistry Letters. 2011; 21: 1997-1999.
9. Aikaterini P, Dorothea K,Christos K, Hadjipavlou-Litina D. Multitarget Molecular Hybrids of Cinnamic acids. Molecules.2014;19: 20197-20226.
10. Korosec B, Sova M, Turk S, Krasevec N, Novak M, Lah L, Stojan J, Podobnik B, Berne S, Zupanec N, Bunc M, Gobec S and Komel R. Antifungal activity of cinnamic acid derivatives involves inhibition of benzoate 4 hydroxylase (CYP53). Journal of Applied Microbiology. 2014; 116: 955-966.
11. Li-Ping G, Cheng-X W, Xian Q D, Xin S, Hu-Ri P, Zhe-S Q. Synthesis and anticonvulsant activity of N-(2-hydroxy ethyl) Cinnamide derivatives. European Journal of Medicinal Chemistry. 2009; 44: 3654-3657.
12. Prateek Sharmaz. Cinnamic acid derivatives, A new chapter of various pharmacological activities. Journal of Chemical and Pharmaceutical research. 2011; 3: 403-423.
13. Bogdashev N N, Tukhovskaya N A , Pogrebnyak A V. Relationship between biological activity and thermodynamic properties of 5, 5-dimethyl-1,3cyclohexanedione derivatives. Pharmaceutical Chemistry Journal. 1998; 32: 31-33.
14. G Rajitha, N Saideepa, P Praneetha. Synthesis and evalution of N-(α-benzamido cinnamoyl) aryl hydrazone derivatives for anti-inflammatory and antioxidant activities. Indian Journal of chemistry. 2011; 50(5): 729-733.
15. Rajitha G, Prasad K.V.S.R.G, Umamaheswari A, Prardhan D, Bharathi K. Synthesis, biological evaluation and molecular docking studies of N-(α- acetamido cinnamoyl) aryl hydrazine derivatives as antiinflammatory and analgesic agents. Medicinal Chemistry Research. 2014; 23(12): 5204-5214.
16. Sowjanya M, Lakshmi narayana M, Sarala Devi T, Anusha S, Rajitha G. Microwave assisted rapid,efficient synthesis and screening of Acyl hydrazone derivatives for antibacterial activity. International Journal of pharmacy and Technology. 2014; 6(1): 6193-6202.
17. Terzioglu N, Gursoy A. Synthesis and anticancer evaluation of some new hydrazine derivatives of 2,6-dimethylimidazo[2,1-b][1,3,4]thiadiazole-5-carbohydrazide. European Journal Medicinal Chemistry. 2003; 38(7): 781-786.
18. Kamath P.R, Sunil, D, Ajees A. A, Pai K.S.R, Biswas S. N'((2-(6-bromo-2-oxo-2H-chromen-3-yl)-1H-indol-3yl)methylene)benzohydrazide as a probable Bcl-2/Bcl-xL inhibitor with apoptotic and anti-metastatic potential. European Journal Medicinal Chemistry. 2016; 14(120): 134-147.
19. Lilia M, Coralia B, Ion B, Carmen C.D, Anamaria H, Otilia B, Petre I, Anca P, Arnaud T, Irina Z. Synthesis and biological activities of some new isonicotinic acid 2-(2-hydroxy-8-substituted-tricyclo[7.3.1.02.7]tridec-13-ylidene)-hydrazides. Bioorganic Medicinal Chemistry. 2015;23(3): 401- 410.
20. G Rajitha, G Nagalakshmi, K Varalakshmi, P Praneetha. Synthesis and Biological evaluation of Novel Cinnamic Acid derivatives for anti-bacterial and antioxidant activities. International Journal Chemical Sciences. 2011; 9(4): 1811-1818.
21. M Soujanya, G Rajitha, A Umamaheswari, K Sudheer Kumar. Synthesis and Biological evaluation and Docking studies of N-(2-benzamido feruloyl) Aryl Hydrazone Analogues. Letters in Drug Design& Discovery. 2018; 15: 875-886.
22. Ghulam F, Nighat A, Muhammad A V, Nazia F, Uzma R M, Mahboob A K, Lubna I and Mehreen L. Synthesis, spectroscopic characterization and pharmacological evaluation of oxazolones derivatives. Journal of Serbian Chemical Society. 2013; 78: 1127-1134.
23. T V MaruthiKumar, P Hanumantha Rao. A novel synthesis of 1-(1-aza-2-arylvinyl)-2-[(1E)-2-arylvinyl]-4-(phenyl methylene)-2-imidazolin-5-ones. Indian Journal of Chemistry. 2005; 44: 1497-1499.
24. Shadomy S, Espinel AA. Manual of Clinical Microbiology. American Society for Microbiology,Washington. D.C. 1980; 647.
25. Horakova L, Ondrejickova O, Bachrata K, Vajdova M. Preventive effects of several antioxidants after oxidative stress on rat brain homogenates. General Physiology Biophysics 2000; 19(2): 195-205.

26. Gulcin I. The antioxidant and radical scavenging activities of black pepper (Piper nigrum) seeds. International Journal of Food Science and Nutrition. 2005; 56(7): 491-499.
27. Marcocci L, Maguire J J, Droy-Lefaix M T, Packer L. The nitric oxide scavenging properties of Ginkgo biloba extract EGb 761. Biochemical and Biophysical Research Communications. 1994; 201(2): 748-755.
28. Jayaprakah G K, Lingamallu J R, Kunnumpurath, K S. Antioxidant activities of flavidin in different in vitro model systems Bioorganic and Medicinal Chemistry. 2004; 12(19): 5141-5146.
29. Molinspiration cheminformatics [homepage on the internet], Available fromhttp://www.molinspiration.com/ cgi-bin/properties/.
30. www.swissadme.ch/27-06-2018.
31. Xiao LW, Yan BZ, Jian-FT, Yu SY, Ruo QC, Fei Z and Hai-Liang Z. Design, synthesis and antibacterial activities of vanillic acylhydrazone derivatives as potential β ketoacyl-acyl carrier protein synthase III (FabH) inhibitors. European Journal of Medicinal Chemistry. 2012; 57: 373-382.
32. Eklund P C, Langvik, O K, Wearna J P Salmi, T O, Willfeor, S M, Sjeoholm R.E. Chemical studies on antioxidant mechanisms and free radical scavenging properties of lignans. Organic and Biomolecular Chemistry. 2005; 3(18): 3336-3347.
33. Benzie I F, Strain, and J J Ferric reducing / antioxidant power assay: direct measure of total antioxidant activity of biological fluids and modified version for simultaneous measurement of total antioxidant power and ascorbic acid concentration. Methods in Enzymology. 1999; 299: 15-27.

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