IJPSR (2018), Volume 9, Issue 12 (Research Article)

INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES AND RESEARCH

Received on 02 April 2018; received in revised form, 05 July 2018; accepted, 27 November 2018; published 01 December 2018

N-[MORPHOLINO (4-NITROPHENYL) METHYL] NICOTINAMIDE AND ITS METAL COMPLEXES: SYNTHESIS AND EXPLORATION OF THEIR BIOACTIVITY

S. Kannan [1, * 2], K. K. Mohamed Ameen [2] and M. Syed Ali Padusha [2]

PG and Research Department of Chemistry [1], L. N. Government College (Autonomous), Ponneri - 601204, Tamil Nadu, India.
PG and Research Department of Chemistry [2], Jamal Mohamed College (Autonomous), Trichy - 620020, Tamil Nadu, India.

Keywords:

Morpholine derivatives, Nicotinamide, Mannich base, Metal complexes, DPPH free radical scavenging and anti-inflammatory activities

Correspondence to Author:
S. Kannan

PG and Research Department of Chemistry, L. N. Government College (Autonomous), Ponneri - 601204, Tamil Nadu, India.

E-mail: sipikannan@yahoo.com

ABSTRACT: Synthetic organic compounds, containing relatively higher number of nitrogen attracting interest in the current medical research due to their utility in chemotherapy. The development of new medicines with more number of nitrogen atoms in their structural framework is an important challenging task for medicinal chemists. The lipophilicity of Mannich bases and their metal complexes enables them to be more potent novel medicines and acquire huge interest among medicinal chemists. We report here the synthesis of novel lead structure of new Mannich base and its complexeswith the variety of transition metals such as Co(II), Mn(II), Ni(II), Cu(II) and Zn(II) and evaluated them for DPPH free radical scavenging and anti-inflammatory activity. As the Mannich base from the combinations 4-nitrobenzaldehyde, morpholine and nicotinamide was not yet reported, the new Mannich base, N-[Morpholino(4-nitrophenyl)methyl]nicotinamide (NMN), was synthesized from this combination. The ligand and its complexes were characterized using various analytical (Chemical assays, Elemental analysis and TLC) and spectral studies (FT-IR, UV-Visible, ^{1}H NMR, ^{13}C NMR, Mass and TGA).

INTRODUCTION: The Mannich base obtained from amines and amides are found to possess increasing lipohilicity [1-3], which results in the enhancement of absorption through bio-membranes [4]. Due to this activity, a widespread pharmaceutical applications [5] including antibacterial [6], anthelmintic [7], antifungal [8], anti-inflamatory [9], antiviral [10] and analgesic [11] properties are prompted the researchers into the synthesis of novel lead structure for the designing of new, potent and less toxic agents, which ideally shorten the duration of therapy.

QUICK RESPONSE CODE

DOI: 10.13040/IJPSR.0975-8232.9(12).5511-21

Article can be accessed online on: www.ijpsr.com

DOI link: http://dx.doi.org/10.13040/IJPSR.0975-8232.9(12).5511-21

Mannich bases can form stable complexes with various transition metals [12-16] and show enhanced biological activities. An important aspect of medicinal chemistry has been a great deal of success in understanding relationship with chemical structure and its biological activity.

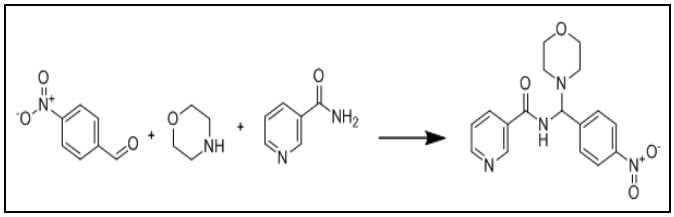
The derivatives of heterocyclic compounds have their own importance due to the good biological activities. Among the wide variety of heterocycles, the compounds bearing nitrogen atom as hetero atom have played an important role in medicinal chemistry, in this context we focused here to synthesis a new compound from the combination of 4-nitrobenzaldehyde, morpholine and nicotinamide as they were not yet reported. The presence of amide moiety as a functional group have strong ability to form metal complexes, so that the disease

causing microorganisms absorb it through bio-membranes easily [17-23]. In continuation of our ongoing research program in the field of synthesis and bioactivity of medicinally important compounds, here we report the synthesis, characterization, DPPH free radical scavenging and anti-inflammatory studies of a Mannich base, N-[Morpholino (4- nitrophenyl) methyl] nicotinamide and its complexes with a variety of transition metals such as Co(II), Mn(II), Ni(II), Cu(II) and Zn(II).

EXPERIMENTAL METHODS: All the chemicals were of AR grade and used without further purification unless otherwise stated. All the aromatic aldehydes were obtained from Avra Synthesis Pvt. Ltd., Hyderabad. Melting points of all the compounds were determined in open capillaries and are uncorrected. The homogeneity of compounds was checked by TLC on a silica gel 'G' coated glass plates.

IR spectra were recorded in KBr on Shimadzu FT-IR 8300 spectrometer at SRM University, Chennai. ^{1}H NMR and ^{13}C NMR spectra were recorded at Varian 400 MHz in Bruker Advance II instruments using DMSO-d6 as a medium and TMS as an internal standard. Mass spectrum was recorded (HPLC+PDA+MS) at SRM University. TGA was carried out using the instrument TGA Q50 V20 13 Buid 39 at CLRI, Chennai. DPPH free radical scavenging and anti-inflammatory activities were carried by the prescribed standard procedure at Mariana Labs, Chennai.

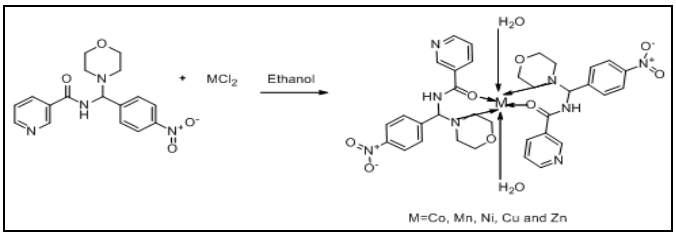
Synthesis of N- [Morpholino (4- nitrophenyl) methyl]nicotinamide (NMN): 4-nitrobenzalde-hyde, morpholine and nicotinamide were taken in 1:1:1 ratio. 1.6 g of p-nitrobenzaldehyde was taken in a round bottomed flask and 10 mL of ethanol was added. To this solution 0.9 mL of morpholine in ethanol was added and stirred well for 15 min by keeping the reaction mixture on a magnetic stirrer.



SCHEME 1: SYNTHESIS OF N-[MORPHOLINO(4-NITROPHENYL)METHYL]NICOTINAMIDE

The solution was made alkaline by adding NaOH pellets. To this solution 1.3 g of nicotinamide in ethanol was added and stirred. Stirring was continued under ice cold condition for about 8 h. The compound thus formed was filtered, washed and recrystallized using ethanol, **Scheme 1**.

Synthesis of Transition Metal Complexes: A solution of 0.1M of MCl_2 (M=Co, Mn, Ni, Cu and Zn) in methanol and 0.2M of N-[Morpholino(4-nitrophenyl)methyl]nicotinamide in ethanol were added to a round bottomed flask and stirred well using magnetic stirrer for an hour, **Scheme 2**. The complex formed was filtered, washed with distilled water and crystallized from absolute alcohol.



SCHEME 2: SYNTHESIS OF METAL COMPLEXES

***In-vitro* Studies:** *In-vitro* studies such as antioxidant and anti-inflammatory were carried outfor the synthesized compound N-[Morpholino (4-nitrophenyl) methyl] nicotinamide and its complexes. The presence of aminoalkyl chain is a key feature shown by number of medicinal agents.

Antioxidant Activity: Oxidation is one of the most important processes, which produce free radicals in food, chemicals, and also in metabolism. Free radicals have an important role in processes of food spoilage, chemical materials' degradation and also contribute to more than one hundred disorders in humans. Antioxidants can significantly delay or prevent oxidation of easy oxidizable substrates even at very low concentrations. The applications of antioxidants are industrially widespread in order to prevent polymers oxidative degradation, auto-oxidation of fats, synthetic and natural pigments discoloration, *etc*. There is an increased interest of using antioxidants for medical purposes in the recent years.

Several methods are used for the estimation of efficiency of synthetic / natural antioxidants, like the ferric reducing antioxidant power (FRAP) assay, *β*-carotene / linoleic acid assay, Rancimat method, inhibition of low-density lipoprotein

oxidation, DPPH assay, *etc*. This method diversity is due to the complexity of the analyzed substrates, often mixtures of dozens of compounds with different functional groups, polarity, and chemical behavior. In this paper the attention is focused on the DPPH assay, which is one of the best-known, frequently employed, and accurate methods. DPPH (1,1-diphenyl-2-picrylhydrazyl) is a stable free radical because of its spare electron delocalization over the whole molecule. The delocalization causes a deep violet color with λ_{max} around 520 nm. When a solution of DPPH is mixed with a substrate acting as a hydrogen atom donor, a stable non-radical form of DPPH is obtained with simultaneous change of the violet color to pale yellow.

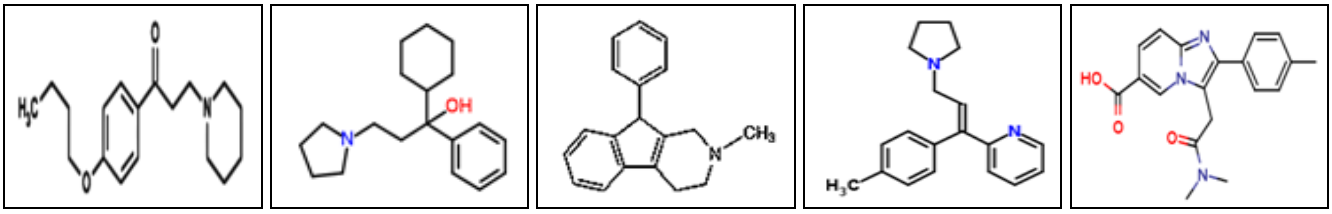
The percentage of antioxidant activity of each substance was assessed by DPPH free radical assay. The measurement of the DPPH radical scavenging activity was performed according to methodology described by Brand-Williams *et al*. The samples were prepared at various concentrations (10, 20, 30, 40 and 50 µg/mL) in methanol and taken in small tubes. They were allowed to react with the stable DPPH radical in methanol solution. The samples were made up to 1 ml using methanol and 1 ml of DPPH in methanol was added to each of the tube. The same solution of DPPH in methanol was used as the control and Butylated Hydroxyanisole (BHA) was used as the reference. The mixture of methanol and sample was served as blank. When DPPH reacts with an antioxidant compound which can donate hydrogen, the DPPH is reduced. The changes in color (from deep violet to light yellow) were read at 517 nm after 30 min of incubation in dark at room temperature using a UV-VIS spectrophotometer. The scavenging activity percentage was determined and expressed as percentage decrease with respect to control values. The experiment was done in triplicate for each substance. The percentage of inhibition was calculated using the following formula.

Effective concentration % = (Control Absorbance – Test Absorbance) / Control Absorbance × 100

Anti inflammatory Activity: Inflammation is a complex process, which is frequently associated with pain and involves occurrences such as: the increase of vascular permeability, increase of protein denaturation and membrane alteration. Protein denaturation is a process in which proteins lose their tertiary and secondary structure by application of external aid, such as strong acid or base, a concentrated inorganic salt, an organic solvent or heat. Most biological proteins lose their biological function when they denatured. Denaturation of protein is a well-documented cause of inflammation. As part of the investigation on the mechanism of the anti-inflammatory activity, ability of some synthesized compounds to inhibit protein denaturation was studied. Many disease conditions and surgical procedures are associated with pain and inflammation. The currently available analgesic and anti-inflammatory agents such as aspirin, diclofenac, indomethacin, ibuprofen, naproxen and others are carboxylic acid derivatives and are associated with ulcerogenic effect.

The current approaches utilizes to mask the ulcerogenic effect of these drugs includes prodrug concept and conversion of carboxylic group to some other functional groups such as amide, ester, aldehyde or ketones. The N-substituted N-[Morpholino (4-nitrophenyl) methyl] nicotinamide and its complexes enjoys some common therapeutic actions which include antibacterial, analgesic and anti-inflammatory. Number of potent medicinal agents consists of aminoalkyl chain in its structure. Many mannich bases, which are identified by the presence of aminoalkyl chain, are in clinical use such as cocaine, dyclonine, tutocaine, tanitidine, phenindamine, triprollidine, amodiaquin, ethacrynic acid, procyclidine, trihexyphenidyl, molindone, zolpidem, fluoxetine and propoxyphene.

A solution of 0.2% (w/v) of BSA was prepared in a Tris Buffer Saline and pH was adjusted to 6.8 using glacial acetic acid. To different concentrations of the sample (1-5 mg/mL), 5 mL of 0.2% w/v BSA was added. The control system was prepared as 5 mL 0.2% (w/v) BSA solution with 50 µL methanol. The test tubes were heated to 72 ºC for 5 min and then cooled to room temperature. The absorbance of these solutions was determined in a UV-Vis spectrophotometer at wavelength of 660 nm. The percentage of inhibition of precipitation (denaturation of the protein) was determined on a percentage basis relative to control.



Dyclonine **Procyclidine** **Phenindamine** **Triprollidine** **Zolpidem**

RESULTS AND DISCUSSION:

Elemental Analysis: The molecular formula of the synthesized compound was proposed as $C_{17}H_{18}N_4O_4$, which was confirmed by the elemental analysis. The results of elemental analyses show 1:2 (metal: Ligand) stoichiometry for all the complexes which confirms the suggested general formula as $[C_{34}H_{40}N_8O_{10}M]Cl_2$. The analytical data of the ligand and their complexes are given in **Table 1**. The high molar conductance of the chelates in DMF supports the electrolytic nature of metal complexes [24-27].

TABLE 1: PHYSICAL PROPERTIES

Compounds	Molecular formula	Molecular weight	Decomposition temperature	Conductance ($\Omega^{-1}cm^2mol^{-1}$in 10^{-3})
NMN	$C_{17}H_{18}N_4O_4$	342	140ºC	--
Mn(II)-NMN	$[C_{34}H_{40}N_8O_{10}Mn]\ Cl_2$	846	154ºC	200
Co(II)-NMN	$[C_{34}H_{40}N_8O_{10}Co]\ Cl_2$	850	156ºC	182
Ni(II)-NMN	$[C_{34}H_{40}N_8O_{10}Ni]\ Cl_2$	850	158ºC	185
Cu(II)-NMN	$[C_{34}H_{40}N_8O_{10}Cu]\ Cl_2$	854	159ºC	164
Zn(II)-NMN	$[C_{34}H_{40}N_8O_{10}Zn]\ Cl_2$	856	160ºC	162

TABLE 1A: ELEMENTAL ANALYSIS

Compounds	Molecular formula	Color	Elemental analysis % Calculated (Found)			
			C	H	N	O
NMN	$C_{17}H_{18}N_4O_4$	Yellowish orange	59.64 (56.8)	5.26 (5.56)	16.37 (16.00)	18.7 (19.00)
Mn(II)-NMN	$[C_{34}H_{40}N_8O_{10}Mn]\ Cl_2$	Reddish brown	48.23 (48.7)	4.72 (4.92)	13.24 (13.62)	18.9 (19.01)
Co(II)-NMN	$[C_{34}H_{40}N_8O_{10}Co]\ Cl_2$	Pale pink	48.00 (48.52)	4.70 (5.02)	13.17 (13.82)	18.8 (19.03)
Ni(II)-NMN	$[C_{34}H_{40}N_8O_{10}Ni]\ Cl_2$	Brown	48.02 (48.5)	4.70 (5.23)	13.18 (13.22)	18.18 (18.33)
Cu(II)-NMN	$[C_{34}H_{40}N_8O_{10}Cu]\ Cl_2$	Green	47.75 (48.33)	4.68 (4.82)	13.10 (13.35)	18.72 (19.22)
Zn(II)-NMN	$[C_{34}H_{40}N_8O_{10}Zn]\ Cl_2$	Yellowing brown	47.64 (49.22)	4.67 (5.02)	13.0 (13.51)	18.68 (19.22)

FT-IR Spectra: The structural relationship among the constituent atoms and group of the synthesized compound and its complexes were established using the data obtained from IR spectroscopy **Fig. 1A – 1F**). IR frequencies corresponding to the respective vibrations are summarized in **Table 2**. The mode of coordination and its' sites were pointed out by comparing the IR spectrum of ligand and its complexes.

The normal ν_{C-H} of alkanes and aromatics are in the range of 3173-2810 cm^{-1}. The characteristic IR band observed at 2963 cm^{-1} is attributed to the ν_{ArC-H}. The band appeared at 2903 cm^{-1} is assigned to νAli_{C-H}. The presence of C=O and C-N has been confirmed by the band observed at 1679 cm^{-1} and 1345 cm^{-1} respectively. The $\nu_{C=O}$ of the ligand in complex was found shifted by 07 to 78 cm^{-1}; indicates the coordination of oxygen atom of carbonyl group of nicotinamidewith the metal ion. The ν_{CNC} of morpholine is lowered by 149 cm^{-1} in the spectra of the complexes suggesting the coordination is through N atom of morpholine. These changes were further advocated by a medium intensity band observed in the range 544 cm^{-1} and 530 cm^{-1} for all the complexes are due to the νM-O and νM-N respectively [28-30]. The broad bands ranging from 3348 to 3559 cm^{-1} confirm the presence OH stretching. IR data concludes that the ligand acts as a bidentate and coordination occurs through N and O atoms to the metal ions.

TABLE 2: CHARACTERISTIC IR BANDS (cm^{-1}) OF NMN AND ITS METAL COMPLEXES

Entry	Compound	Band assignment, cm^{-1}							
		υH_2O	**υ Ar-H**	**υAli-CH**	**υC=O**	**υC-N-C**	**υC-O-C**	**υM-N**	**υM-O**
1	NMN	3364	2963	2851	1679	1273	1109	--	--
2	Mn(II)-NMN	3422	2981	2929	1601	1128	1073	445	519
3	Co(II)-NMN	3558	2928	2858	1672	1124	1074	453	530
4	Ni(II)-NMN	3385	2982	2931	1677	1125	1075	455	533
5	Cu(II)-NMN	3349	2982	2859	1603	1126	1074	474	544
6	Zn(II)-NMN	3348	2981	2932	1641	1126	1074	486	530

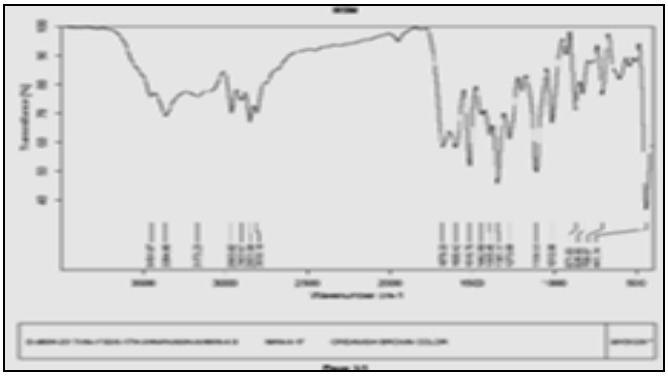


FIG. 1A: FT-IR SPECTRUM OF NMN

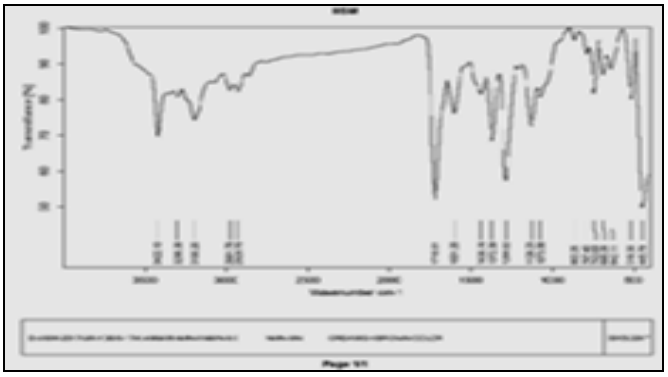


FIG. 1B: FT-IR SPECTRUM OF Mn-NMN

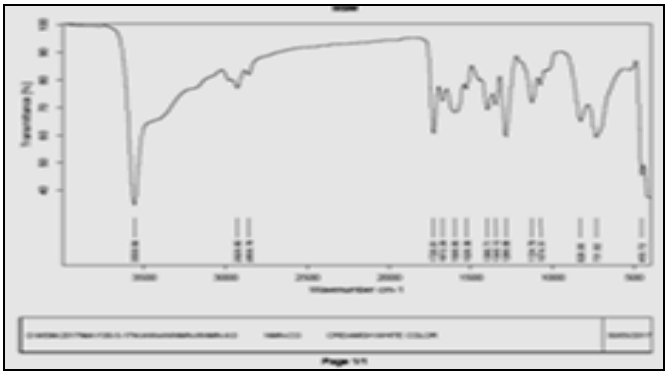


FIG. 1C: FT-IR SPECTRUM OF Co-NMN

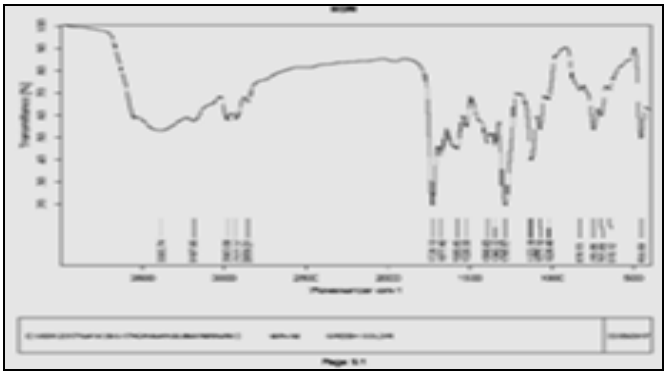


FIG. 1D: FT-IR SPECTRUM OF Ni-NMN

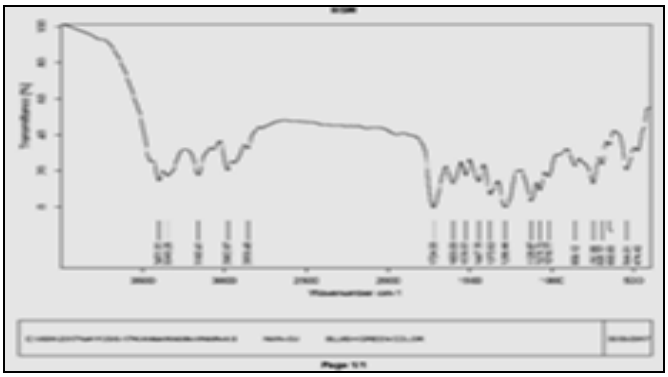


FIG. 1E: FT-IR SPECTRUM OF Cu- NMN

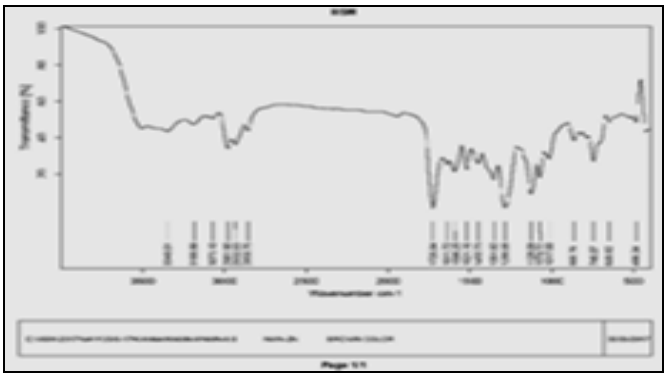


FIG. 1F: FT-IR SPECTRUM OF Zn-NMN

Electronic Spectra: The UV-visible spectra **Fig. 2A – 2E** values of the ligand and its complexes are adding still more evidences on the structural investigations. The electronic spectral measure-ments were used for assigning the structural relationships among the constituent groups of metal complexes based on the position and number of d-d transitions.

The electronic absorption spectra of Co(II), Mn(II), Ni(II), Cu(II) and Zn(II) complexes of NMN were recorded at room temperature using 10^{-3} M solution of the complex prepared using DMSO as solvent. It was recorded in the range of 250-900 nm. The intensity of absorption and its corresponding electronic transitions [31-34] are summarized in **Table 3**.

TABLE 3: UV-VIS SPECTRAL AND MAGNETIC DATA

Entry	Compounds	Absorption		Transition	Magnetic moment (BM)	Geometry
		nm	cm^{-1}			
1	Mn(II)-NMN	259	38,610	$^6A_1g \rightarrow {}^4E_1g$,	5.82	Octahedral
		360	27,777	$^6A_1g \rightarrow {}^4T_2g$		
2	Co(II)-NMN	266	37594	$_4T_1g(F) \rightarrow {}_4T_2g(P)$	3.84	Octahedral
		320	31250	$_4T_1g(F) \rightarrow {}_4A_2g(F)$		
3	Ni(II)-NMN	254	39370	$\pi \rightarrow \pi^*$	2.86	Octahedral
		288	34722	$^3A_2g(F) \rightarrow {}^3T_1g(F)$		
		306	32679	$^3A_2g(F) \rightarrow {}^3T_1g(P)$		
		306	32679	$^3A_2g(F) \rightarrow {}^3T_1g(P)$		
4	Cu(II)-NMN	250	40000	$\pi \rightarrow \pi^*$	2.74	Octahedral
		302	33112			

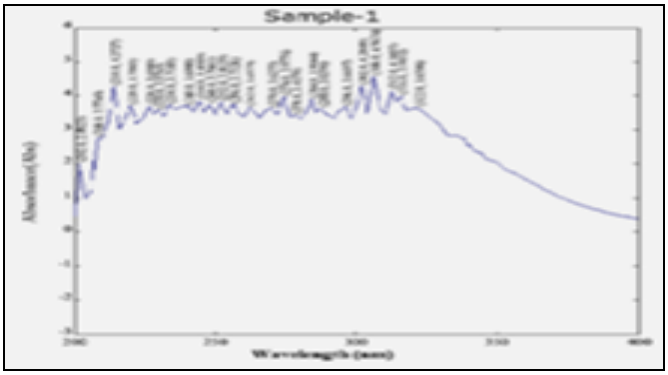


FIG. 2A: UV SPECTRUM OF NMN

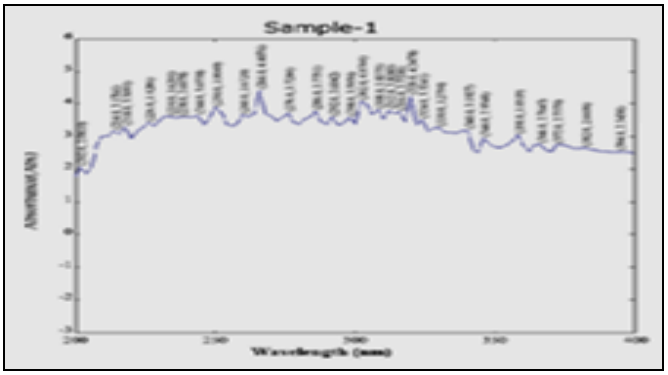


FIG. 2B: UV SPECTRUM OF Co-NMN

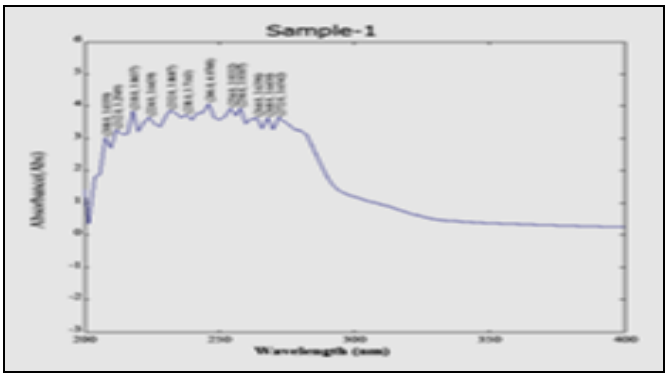


FIG. 2C: UV SPECTRUM OF Mn-NMN

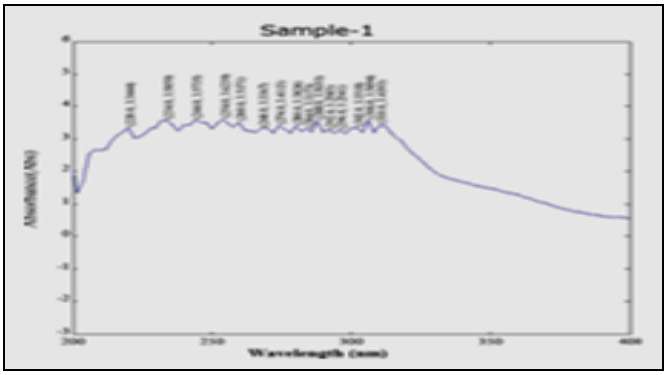


FIG. 2D: UV SPECTRUM OF Ni-NMN

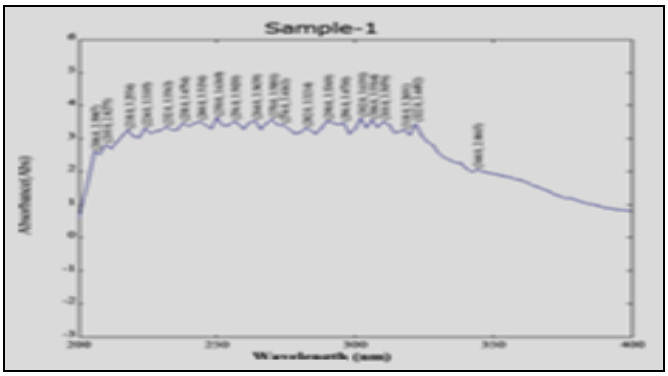


FIG. 2E: UV SPECTRUM OF Cu- NMN

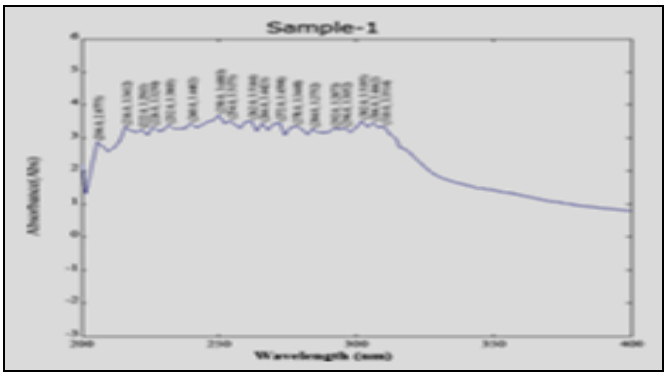


FIG. 2F: UV SPECTRUM OF Zn-NMN

^{1}H NMR Spectrum: ^{1}H NMR spectrum of the ligand was recorded at SRM University, in DMSO-d6 medium using TMS as an internal standard and the spectrum is shown in **Fig. 3**. The results obtained from ^{1}H NMR spectrum are used to find out the number of protons and their chemical environments. The structural relationship among the 18 protons was identified from ^{1}H NMR data.

The multiplets obtained from 9.02 to 7.50 ppm are assigned to the protons of nicotinamide ring. The triplets appeared at 3.57 ppm and 2.65 ppm are corresponding to CH_2-O-CH_2 and CH_2-N-CH_2 of morpholine ring respectively. The aromatic protons of phenyl ring in O_2N-Ph-CH ortho to –CH group appeared as a doublet at 7.59 ppm whereas the aromatic protons of the same phenyl ring meta to –CH group appear as a doublet at 8.14 ppm. The signals found at 8.03 ppm and 6.05 ppm are corresponding to –NH and –CH proton respectively. The ^{1}H NMR experimental data was supported by the data obtained from the ChemDraw 16.0 software.

^{13}C NMR Spectrum: ^{13}C NMR spectrum of the ligand was recorded at SRM University, in DMSO-d6 medium using TMS as an internal standard and the same is shown in **Fig. 4**. The results obtained from ^{13}C NMR are helped to find out the number of carbons and their chemical environments. The structural relationship among the 17 carbons was also established. The peaks observed from 124.3 ppm to 154.4 are assigned to carbon atoms of nicotinamide. The peak appeared at 167.8 ppm is attributed to carbonyl carbon. The aromatic carbons of phenyl ring in O_2N-Ph-CH ortho to –CH group appeared at 130.1 ppm whereas the aromatic carbons of the same phenyl ring meta to –CH group appeared 127.6 ppm. The other carbons of the same ring are observed at 147.5 ppm and 142.2 ppm. The peaks appeared at 66.7 ppm and 49.5 ppm is due to CH_2-O-CH_2 and CH_2-N-CH_2 of morpholine ring respectively. The aliphatic carbon was appeared at 87.1 ppm.

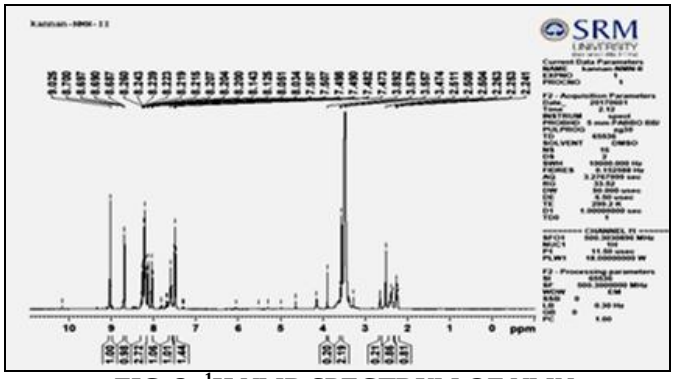


FIG. 3: ^{1}H NMR SPECTRUM OF NMN

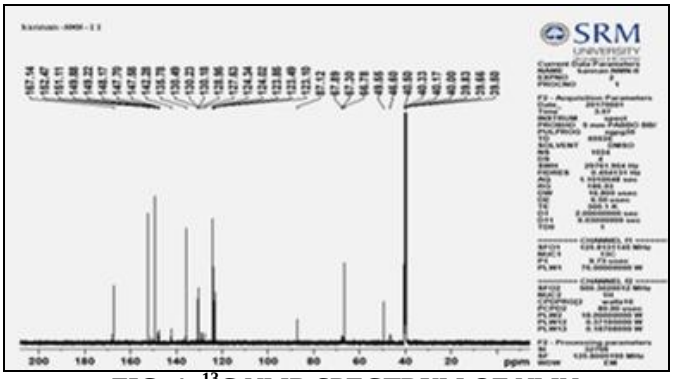


FIG. 4: ^{13}C NMR SPECTRUM OF NMN

Mass Spectroscopy (LC-MS): The Mass spectrum of NMN was recorded at SRM University, Chennai, by electro ionization mode, **Fig. 5**. The spectrum shows a molecular ion peak at m/z = 341, which confirms the assigned molecular mass to the Mannich base, N-[Morpholino(4-nitrophenyl)-methyl]nicotinamide. The intensesignal at m/z = 100 is due the final fragment after the removal of nitro phenyl and nicotinamide units.

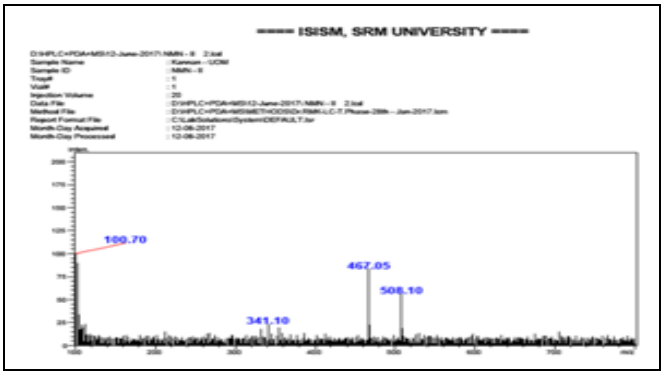
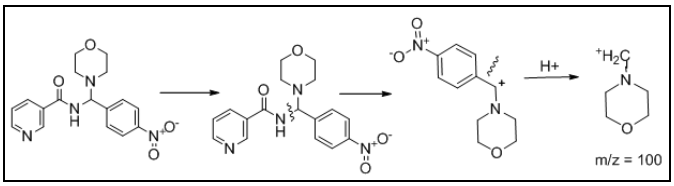


FIG. 5: MASS SPECTRUM OF NMN



SCHEME 3: FRAGMENTATION SCHEME PROPOSED FOR NMN

Thermal Analysis (TGA): The TG curve of the representative complex [$C_{34}H_{40}N_8O_{10}Mn$] Cl_2 was recorded in the temperature range of 0 - 800 ºC and is shown in **Fig. 6**. The decomposition of the complex is completed in five steps. In the first step, an initial weight loss of 5.41% is observed from 0 to 118 ºC corresponding to the loss of one nitro group. Second step shows weight loss of 11.02% attributed to the loss of two morpholine units. Third step is due to the loss of two nicotinamide units. The other fragments such as water molecules and chlorine atoms are lost subsequently. It is decomposed completely at 605.86 ºC. These

observations are supported by the other analytical and spectral studies.

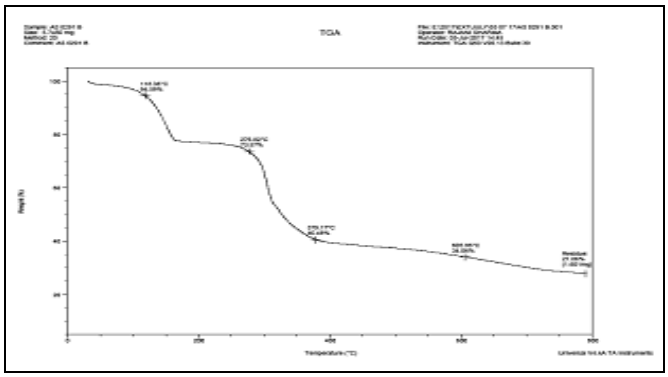


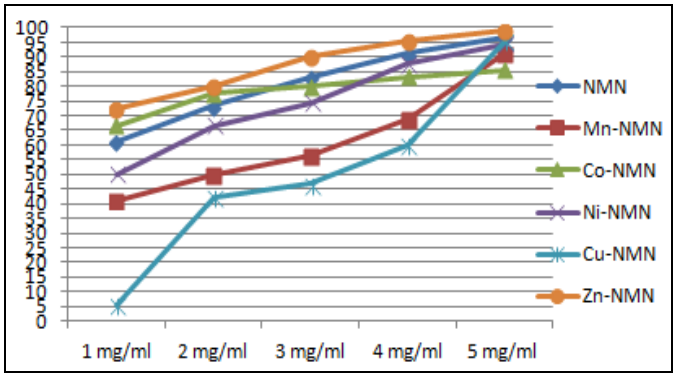
FIG. 6: TGA CURVE OF Mn-NMN

Antioxidant Activity: DPPH radical scavenging activity is one of the methods used most widely for screening the antioxidant activity of drugs. The **Table 4** shows the antioxidant activities of the synthesized mannich base ligand and its metal complexes at varying concentrations.

The DPPH assay shows free radical scavenging activity of the compounds increases with increasing the concentration, **Fig. 7A – 7F**. The highest DPPH scavenging activity was observed in the Zn-NMN (98.89% at 5 mg/mL) followed by NMN(96.67% at 5 mg/mL), Cu-NMN(95.56% at 5 mg/mL), Ni-NMN(94.44% at 5 mg/mL), Mn-NMN(91.11 % at 5 mg/mL) and Co-NMN(85.56 % at 5 mg/mL). The order of DPPH free radical scavenging activity is as follows: Zn-NMN > NMN > Cu-NMN > Ni-NMN >Mn-NMN > Co-NMN **Graph 1**. The presence of more electron donating groups on the ligand enhances the profound antioxidant activity. It is believed that the hetero atoms present in morpholine moiety and nicotinamide units are responsible for effective antioxidants by scavenging radicals. The hetero atoms combined with an amide group may also increase the antioxidant activity of ligand and its complexes [35].

TABLE 4: ANTIOXIDANT ACTIVITY OF NMN AND ITS COMPLEXES

Sample	% Inhibition					EC_{50} (mg/mL)
	1 mg/mL	2 mg/mL	3 mg/mL	4 mg/mL	5 mg/mL	
NMN	61.11	73.33	83.33	91.11	96.67	<1
Mn-NMN	41.11	50	56.67	68.89	91.11	2
Co-NMN	66.67	77.78	80	83.33	85.56	<1
Ni-NMN	50	66.67	74.44	87.78	94.44	1
Cu-NMN	5.56	42.22	46.67	60	95.56	2.99
Zn-NMN	72.22	80	90	95.56	98.89	<1



GRAPH 1: ANTIOXIDANT ACTIVITY OF NMN AND ITS COMPLEXES

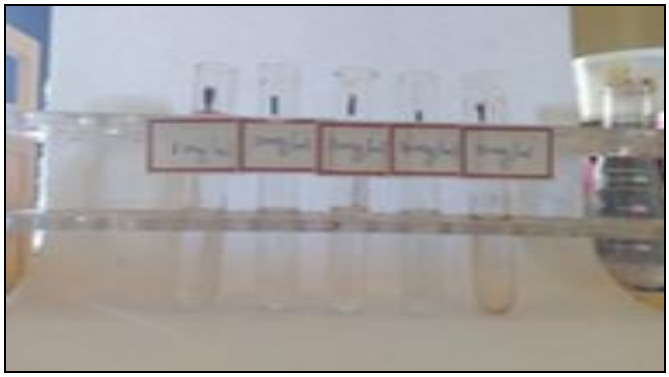


FIG. 7A: DPPH ACTIVITY OF NMN

FIG. 7B: DPPH ACTIVITY OF Mn-NMN

FIG. 7C: DPPH ACTIVITY OF Co-NMN

FIG. 7D: DPPH ACTIVITY OF Ni-NMN

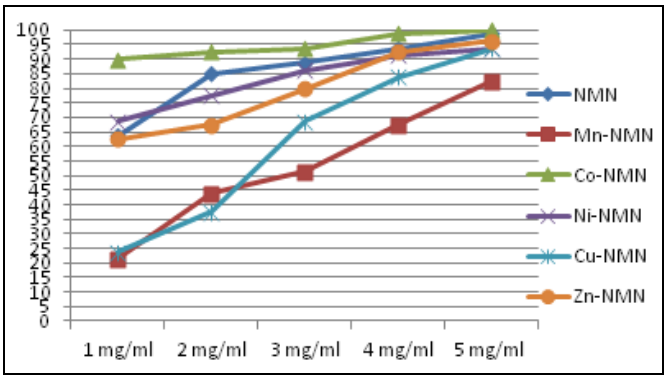
FIG. 7E: DPPH ACTIVITY OF Cu-NMN

FIG. 7F: DPPH ACTIVITY OF Zn-NMN

Anti-inflammatory Activity: The synthesized mannich base ligand and its complexes were subjected to screen their potential towards anti-inflammatory activity, **Fig. 8A – 8E**. The **Table 5** shows the anti-inflammatory activities of the synthesized mannich base ligand and its metal complexes at varying concentrations. It has been found that the activity of above compounds increases with increasing the concentration, **Graph 2**. The highest anti-inflammatory activity was observed in the complex Co-NMN (100% at 5 mg/ml) followed by NMN (98.75% at 5 mg/mL), Zn-NMN (96.255% at 5 mg/mL), Ni-NMN (93.75% at 5 mg/mL), Cu-NMN (93.75% at 5 mg/mL), and Mn-NMN (82.50% at 5 mg/mL). The effectiveness of anti-inflammatory activity is increased in following order: Co-NMN > NMN > Zn-NMN > Ni-NMN > Cu-NMN >Mn-NMN.

TABLE 5: ANTI-INFLAMMATORY ACTIVITY OF NMN AND ITS COMPLEXES

Sample	% Inhibition					EC_{50} (mg/mL)
	1 mg/mL	2 mg/mL	3 mg/mL	4 mg/mL	5 mg/mL	
NMN	63.75	85	88.75	93.75	98.75	<1
Mn-NMN	21.25	43.75	51.25	67.5	82.5	2.78
Co-NMN	90	92.5	93.75	98.75	100	<1
Ni-NMN	68.75	77.5	86.25	91.25	93.75	<1
Cu-NMN	23.75	37.5	68.75	83.75	93.75	2.98
Zn-NMN	62.5	67.5	80	92.5	96.25	<1



GRAPH 2: ANTI-INFLAMMATORY ACTIVITY OF NMN AND ITS COMPLEXES

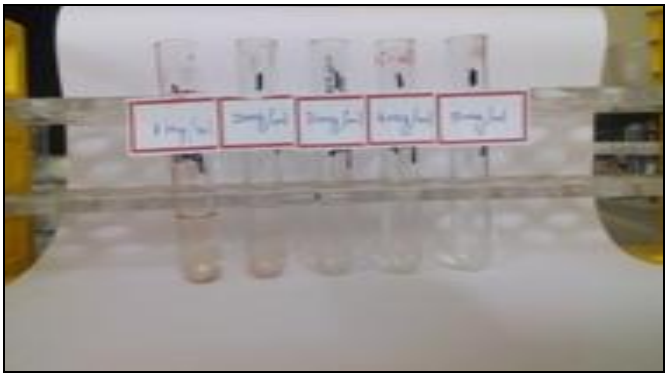


FIG. 8A: ANTI-INFLAMMATORY ACTIVITY OF NMN

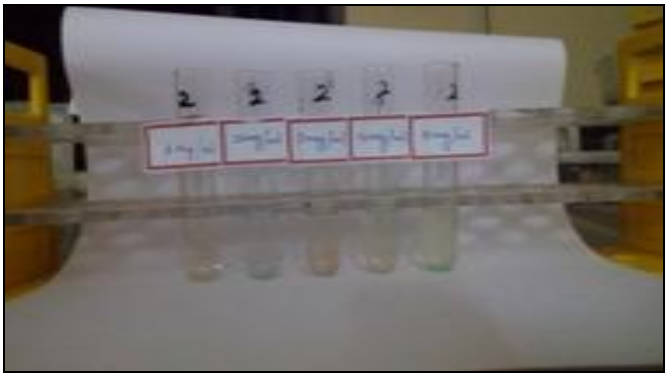


FIG. 8B: ANTI-INFLAMMATORY ACTIVITY OF Mn-NMN

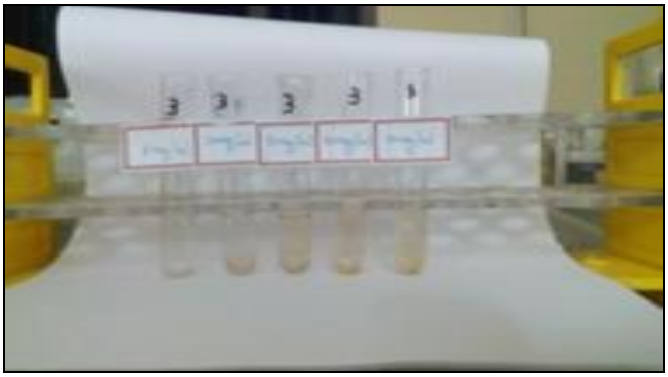


FIG. 8C: ANTI-INFLAMMATORY ACTIVITY OF Co-NMN

FIG. 8D: ANTI-INFLAMMATORY ACTIVITY OF Ni-NMN

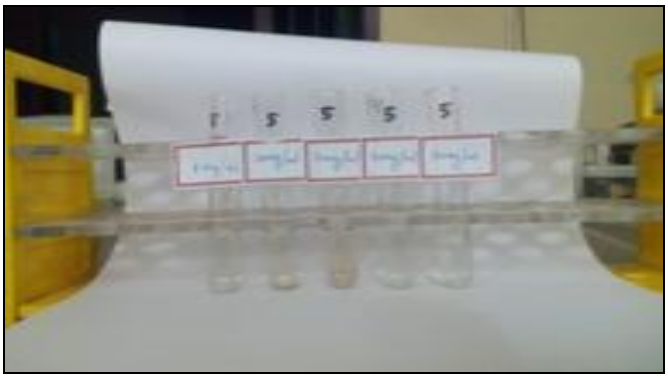


FIG. 8E: ANTI-INFLAMMATORY ACTIVITY OF Cu-NMN

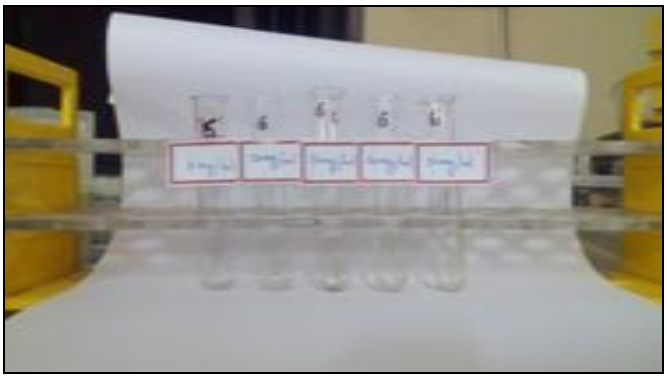


FIG. 8F: ANTI-INFLAMMATORY ACTIVITY OF Zn-NMN

CONCLUSION: In this study a Mannich base and its complexes have been synthesized, characterized and evaluated for antioxidant and anti-inflammatory activity. The structures of all the newly synthesized compounds were confirmed by the suitable analytical (Chemical tests, Elemental analysis and TLC) and spectral studies (FT-IR, UV-Visible, ^{1}H NMR, ^{13}C NMR, Mass and TGA).

The spectral results were concluded the structures of new compound NMN and their complexes. Hence, *in-vitro* studies such as antioxidant and anti-inflammatory were carried out for the synthesized compound N-[Morpholino(4-nitrophenyl) methyl] nicotinamide and its complexes. They have shown excellent antioxidant and anti-inflammatory activities.

High potentiality against the hazardous bioprocesses of the above said compound and its complexes are due to the presence of more hetero atoms in their structures. It was enhanced further due to the presence of electron releasing amide linkage in it. In both the activity studies, the lowest effectiveness was found at 82.5, which is highly potential comparing the other drugs reported earlier. The complexes prepared with N-[Morpholino (4-nitrophenyl)methyl]nicotinamide, derived from the combination of 4-nitrobenzaldehyde, morpholine and nicotinamide could reasonable be used as an effective drug for antioxidant and anti-inflammatory activity. These findings could also be of commercial interest to both pharmaceutical companies and research institutes in designing and developing new drugs.

ACKNOWLEDGEMENT: With much reverence, we offer our hearty thanks to the University Grants Commission, SERO, Hyderabad, for sponsoring the project under the scheme "Minor Research Project". (Ref. no. MRP-5197/14 (SERO), UGC, SERO, Hyderabad).

CONFLICT OF INTEREST: The authors declare no conflict of interest.

REFERENCES:

1. Kannan S, Ali S and Padusha M: Int. J. ChemTech Research 2017; 10 (6): 770-783.
2. Pishawikar SA and More HN: Arabian Journal of Chemistry 2013; 10: 16.
3. Malhotra M, Sharma R, Sanduja M, Kumar R, Jain E and Deep A: Synthesis, characterization and evaluation of Mannich bases as potent antifungal and hydrogen peroxide scavenging agents. Acta Poloniae Pharmaceutica: Drug Research 2012; 69(2): 355-361.
4. Gamal El-Din AR, Hatem AS and Gamal MG: Bio. org. Med. Chem 2009; 17: 3886.
5. Alhadi A, Shaker SA, Yehye WA, Ali HM and Abdullah MA: Bull. Chem. Soc. Ethiopia 2012; 26(1): 95.
6. Rehman M, Imran M, Arif M and Farooq: Science Journal of Chemistry 2013; 1(5): 56-66.
7. Chakkaravarthi K, Gokulakrishnan K, Suman T and Tamilvendan D: International Journal of Pharmacy and Pharmaceutical Sciences 2013; 6(1): 492-495.
8. Kaur A and Kapoor A: Scholars Research Library, Der Pharmacia Letter 2016; 8(12): 157-165.
9. Priya D, Srimathi R and Anjana GV: Asian Jounal of Pharmaceutical and Clinical Research 2018; 11(2): 407-409.
10. Periyasamy S, Dhan RL and Christophe P: Int. J. Pharm. Sci. Res 2010; 1: 105-109.
11. Jesudason EP, Sridhar SK and Padma J: Eur. J. Med. Chem 2009; 44: 2307-2312.
12. Marijana H, Kristina S and Sandra K: Eur. J. Med. Chem 2011; 46: 2274-2279.
13. Muruganandam L and Balasubramanian K: Int. J. Res. Chem. Environ 2013; 3: 113-119.
14. Michael K: Polyhedron 2013; 53: 295.
15. Chandrasekaran T, Suresh M, Ahamed FM, Ali S and Padusha M: Pelagia Research Library, Der Chemica Sinica 2014; 5(5): 81-90.
16. Cummings TF and Shelton JR: J Org Chem 1960; 25: 419.
17. Patel PK and Patel PD: Int. J. ChemTech Res. 2010; 2: 1147.
18. Pandeya SN and Shriram D: Eur J Phar Sci 1999; 9: 25-31.
19. Joshi S and Khosla D: Bio-Org and Med Chem 2004; 12: 571-576.
20. Chodosh LA, Fire A, Samuels M and Sharp PA: J Biol Chem 1989; 264(4): 2250-2257.
21. Muthukumar C, Valarselvan S, Sabastiyan A, Subramanian M and Shanmugavadivel M: Intl. J. Inorg and Bioinorg. Chem 2014; 4: 23-28.
22. Radha S, Mothilal KK and Thamaraichelvan A: Journal of Chemical and Pharmaceutical Research 2016; 8(8): 202-211.
23. Emmanuel J: Journal of Applied Chemistry 2013; 5(3): 2278-5736.
24. Sathya D, Kumaran SJ and Jayachandramani N: Research Journal of Pharmaceutical, Biological and Chemical Sciences 2012; 3(2): 905.
25. Mohan GR, Reddy NY and Kumar BV: Int. J. of Applied Bio and pharmaceutical Technology 2013; 4(1): 38-46.
26. Alaghazh MA, Bayoumi HA, Ammar YA and Aldhlmani SA: J. Molec Struct 2013; 1035: 383.
27. Malhotra M, Arora M, Samad A, Sahu K, Phogat P, Journal of the Serbian Chemical Society 2012; 77(5): 589-597
28. Selvam P: Int J of Pharm Sci and Res 2010; 1(9): 105-119.
29. Anbu S, Kandaswamy K, Moorthy SP, Balsubramanian M, Ponnuswamy MN: Polyhedran 2009; 28: 49.
30. Malhotra M, Sharma R, Sanduja M, Kumar R and Jain E: Acta Poloniae Pharmaceutica: Drug Research 2012; 69(2): 355-361.
31. Lukose G, Mohanan K, Saju S and Rahim S: J. Che and Pharm. Research 2013; 5(5): 241.
32. Sabastiyan A and Suvaikin YM: Pelagia Research Library, Advances in Applied Science Research 2012; 3(1): 45.
33. Shivananda MK and Prakash SP: Journal of Chemical and Pharmaceutical Research 2011; 3(2): 303-307.
34. Pishawikar SA and More HN: Arabian Journal of Chemistry 2013, 10: 10, 016.
35. Chakkaravarthi K, Gokulakrishnan K, Suman T and Tamilvendan D: Int. J. of Pharmacy and Pharmaceutical Sciences 2014; 6(1): 492-495.

How to cite this article:
Kannan S, Ameen KKM and Padusha MSA: N-[Morpholino(4-nitrophenyl)methyl]nicotinamide and its metal complexes: Synthesis and exploration of their bioactivity. Int J Pharm Sci & Res 2018; 9(12): 5511-21. doi: 10.13040/IJPSR.0975-8232.9(12).5511-21.