iJRASET
International Journal For Research in Applied Science and Engineering Technology



INTERNATIONAL JOURNAL FOR RESEARCH

IN APPLIED SCIENCE & ENGINEERING TECHNOLOGY

Volume: 10 **Issue:** I **Month of publication:** January 2022

DOI: https://doi.org/10.22214/ijraset.2022.40137

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International Journal for Research in Applied Science & Engineering Technology (IJRASET)
ISSN: 2321-9653; IC Value: 45.98; SJ Impact Factor: 7.538
Volume 10 Issue I Jan 2022- Available at www.ijraset.com

Photo Induced Reactions by Led-Power Reactor for No Photo release from Nitrosyl Metallic Complex

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Abstract: ***We have developed a simple, low-cost LED lamp reactor that was used efficiently to investigate the photochemistry reactivity of nitrosyl complex. It was possible to ratify the proposal for a photoinduced isomerization of nitrito-N (NOO$^-$) ligand for nitrito-O (ONO$^-$) and nitrosyl-N (NO$^+$) for nitrosyl-O (ON$^+$), in the cis-[Ru(NO)(NO$_2$)(bpy)$_2$](PF$_6$)$_2$ complex, as well as the photoresolution of NO in the cis-[Ru(NO)(NO$_2$)(phen)$_2$](PF$_6$)$_2$ complex. For the complex cis-[Ru(NO)(am)(phen)$_2$](PF$_6$)$_4$ photoreaction investigation raised the proposal of isomerization and photoliberation reactions of NO, which encourages the investigation of this dye system with potential use in therapy Photodynamic.***

Keywords: metal nitrosyls, nitric oxide, metal complex, photochemistry by LED, infrared spectroscopy.

I. INTRODUCTION

Metal nitrosyls can be represented as $[MNOL_n]^{q+}$, (where L are coligands around MNO moiety) and they show variable geometries, coordination numbers and electronic and redox properties [1-6]. The reactivity of the coordinated NO can be modulated by chance ligands, indeed, these compounds was shown to be good candidates for ontrolled release of nitric oxide [2, 4, 7].

Photochemistry and photophysics are remarkable in metal nitrosyls most of them when irradiated with UV light [1, 2, 5, 8-11]. The tuning of electronic properties and thus NO-photolabilization has been explored by several research groups as a strategy for controlled NO release including potential biomedical applications such as in photodynamic therapy (PDT) [12].

Nitrosyl photochemical induced linkage isomers, or metastable states, from MNO (GS), are known for a long time [13, 14]. These isomers are generally referenced as metastable state 1, (MS1, M-ON) and metastable state 2 (MS2, M-□^{2}-NO) [14]. The optically induced refractive-index changes accompanying the formation of metastable states make these compounds good candidates for holographic recording materials and non-linear optics applications [15-17]. Additionally, it has been proposed that linkage isomerism may play an important role in NO photolabilization [18].

Due to very short live times of metastable states at room temperature, most of the knowledge came from low temperature infrared (IR) spectroscopy, differential scanning calorimetry (DSC) and X-ray and neutron diffraction photocrystallographic studies [14].

Among metal nitrosyls, iron and ruthenium are by far the most investigated complexes regarding NO photoinduced linkage isomerization [14]. The IR spectroscopy seems to be the most common and suitable technique to follow these isomerizations. The □(NO) in GS, MS1 and MS2 are fairly different and generally shifts to lower energy going from GS to MS2 [14]. For instance, in $Na_2[Fe(CN)_5NO]$ (SNP) [18], □(NO) for GS, MS1 and MS2 are at 1960, 1835 and 1664 cm^{-1} (□$_{irr}$ = 514.5 nm, T< 100 K), respectively. In addition, theoretical calculations for GS, MS1 and MS2 including coordinated NO stretching frequencies are available for iron and ruthenium nitrosyls and agrees well with the energy lowering mentioned [19,20]. Ruthenium nitrosyl complexes such as $K_2[RuCl_5NO]$ and $K_2[Ru(NO_2)_4(NO)(OH)]$ shows photoinduced isomerizations with MS1 and MS2 metastable states being detected [20-22] while only MS1 was reported for $[Ru(NH_3)_4(NO)(OH)]Cl_2$ [22]. The $[Ru(NO)(NO_2)(bpy)_2](PF_6)_2$ presents an additional challenge. It was reported by Kovalevsky and coworkers [14] that not only nitrito-*N* to nitrito-*O* isomerism linkage is observed (at 200 K) but also nitrosil (GS) to isonitrosyl (MS1) linkage isomerism is observed (only at 90 K). This latter transformation takes place by transference of oxygen atom from NO_2 ligand [14]. Bitterwolf, based on IR spectroscopy changes, have reported the formation of MS2 state for $[Ru(bpy)_2(NO)Cl](PF_6)_2$ in ionic liquid frozen matrix at ca. 90 K upon light irradiation of 300 nm < □$_{irr}$ < 400 nm. Not only GS to MS2 isomerization was observed but also NO loss [23]. The NO photolabilization in solid-stated matrix has also been reported for other nitrosyl metal complexes [24, 25].

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In the view of growing importance of metal nitrosyls as NO donors, their photochemical and photophysical properties regarding NO release and/or formation of metastable isomers, here we devised an effective and relatively inexpensive photoreactor in order to evaluate the interference of light in nitrosyls complexes. The efficiency of the system can be certified by means of results presented here for photoreactions in solid state (KBr plates) at room temperature induced by low emission diode (LED) light source of different wavelengths for some nitrosyls ruthenium and the iron SNP complexes. These photoreactions were performed for the complexes only or in the presence of different NO-scavengers such as $[Ru^{III}(OH_2)(edta)]$ or Mioglobyn. Using this simple approach, a variety of reactions involving nitrosyl complexes were investigated, including donor to scavenger NO transfer.

II. EXPERIMENTAL

A. Chemicals and Reagents

Ruthenium trichloride ($RuCl_3$. $3H_2O$), 2,2_-bipyridine (bpy), 1,10-phenathroline (phen), methylene blue (am), new methylene blue (nam), ammonium hexaphuorofosfate $Na_2[Fe(CN)_5(NO)]$ and $[Ru(Hedta)(OH_2)]$ were purchased as high purity reagents from Sigma-Aldrich Chemicals. Acetone and ethanol (Aldrich) grade P.A were used as supplied. Doubly distilled water was used throughout the work.

B. Synthesis of Ruthenium Complexes

The complexes of interest were synthesized using $RuCl_3.nH_2O$ as the starting material, *cis*-$[RuCl_2(X\text{-}Y)_2]$ (X-Y= bpy or phen) and *cis*-$[Ru(NO_2)_2(X\text{-}Y)_2](PF_6)_2$ were intermediates, until obtaining the target complexes, *cis*-$[Ru(NO)(dye)(X\text{-}Y)_2](PF_6)_4$. (dye = am or nam) [26, 27].

The $[Ru(NO)(NO_2)(X\text{-}Y)_2](PF_6)_2$ and *cis*-$[Ru(NO)(dye)(X\text{-}Y)_2](PF_6)_4$ complex [27] was prepared by dissolving 0.05 g (0.094 mmol) of $[RuCl_2(X\text{-}Y)_2]$ in ethanol/water (5mL/5mL) under argon atmosphere and it was heated at 55 ^oC. after 15 minutes 0.150 g (2.27 mmol) of $NaNO_2$ dissolved in ethanol/H_2O (2.5 mL/2.5 mL) was added, for nitrito complex. For dye complexes was use and 0.030 g (0.094 mmol) of am or 0.035 (0.094 mmol) of nam and 0.033 g (0.0187 mmol) for potassium nitrite. For all systems the mixture was kept under stirring and argon atmosphere at 55^oC for 2 h followed by addition 100 mg of NH_4PF_6 (0.61 mmol) and 1 mL of solutions HPF_6 0.1 mol L^{-1}. The resulting green precipitate was collected by filtration. Yield was better than 85%. The *cis*-$[Ru(NO)Cl(phen)_2](PF_6)_2$ were prepared by published procedure reported by Godwin and Meyer [26].

C. LED Device

The proposed photoreactor is a mechanical and optical device capable on maintain an adjustable and constant illumination of electrochemical samples using LED lamps, thus, allowing positioning adjustments, as well as different illumination configurations.

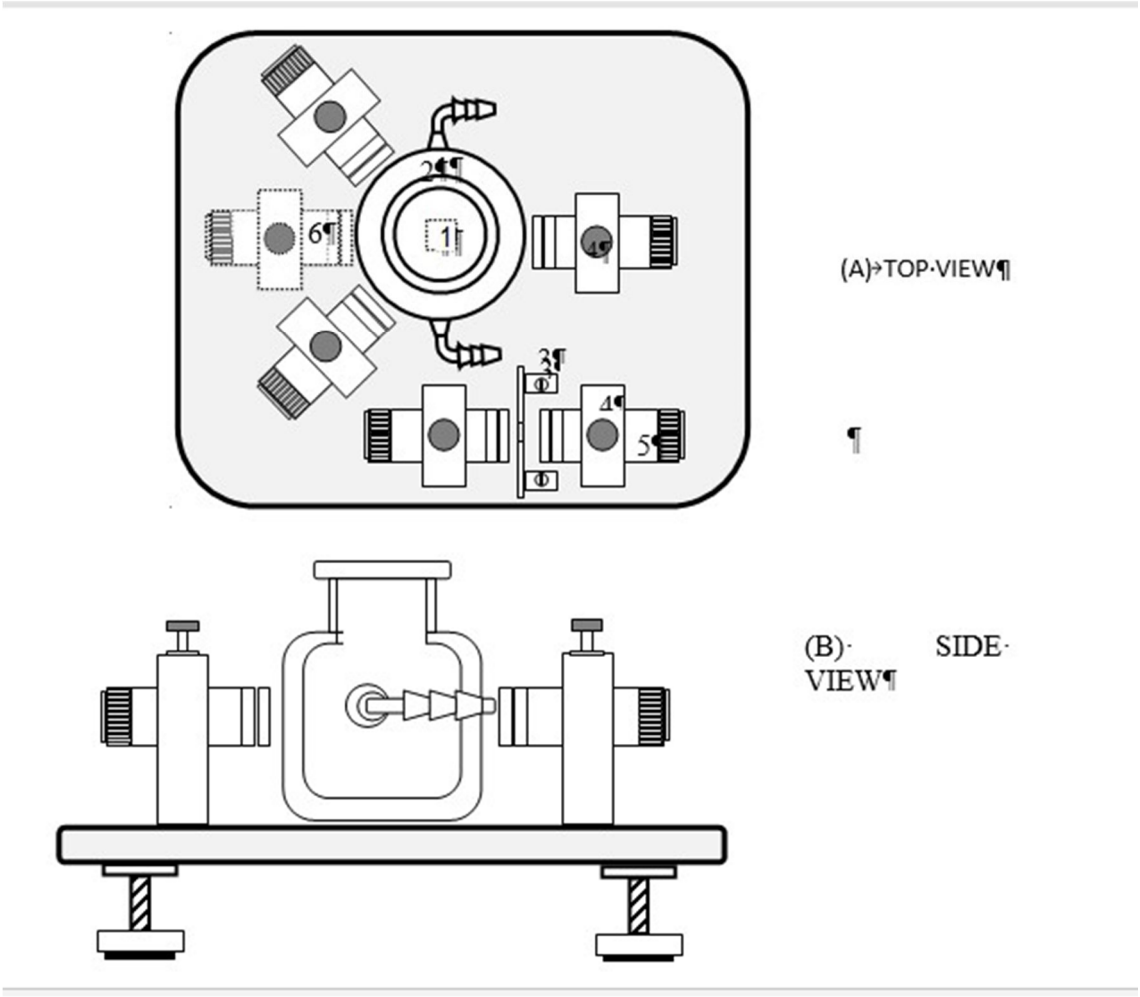
The graphic representation 1 shows the mechanical apparatus capable on operating in three different manners: (i) as a large volume (5 to 20 mL electrochemical cell) three LED lamps illuminator; where the lamps are placed at 120^o respect to the center of the reactor, (ii) as IR-pellets two LED lamps illuminator, irradiating both faces of the pallets with; this functionality allows sample irradiation before or in-betweens IR spectroscopy characterization, (iii) the third illumination mode concerns small (3 mL) quartz spectroscopic cuvettes irradiation using two LED lamps as well.

Nine single 5 mm LED´s are mounted as LED lamps (see graphic representation 2), inside an aluminum housing (d), conferring robustness and simplicity to the irradiation operation, especially on assembling and wavelength selection matters. The changes on light frequency are done replacing; by hand; high efficiency, diverse colors, LED lamps. That way, with very low coast, it is possible irradiate samples at 355, 365, 407, 475 nm, for example. The LED generates a quite narrow Gaussian emission beam for each wavelength, 20 nm wide, with no or very small heating emission.

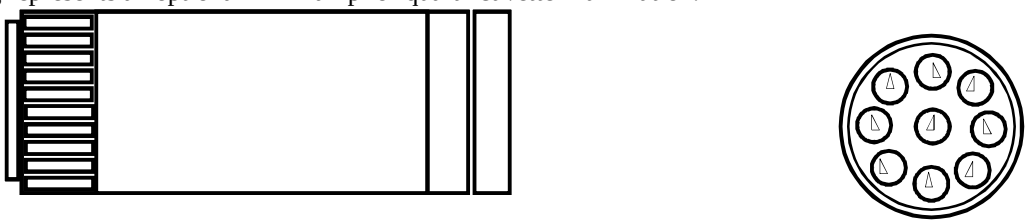
For a given LED and for experimental control sake, its output light intensity can be easily determined with a calibrated photodiode. The procedure is quite simple and could be used for single or multiple LED lamp ensembles. Initially attach the LED lamp to a rigid optical rod, place the photodiode to the distance of interest, fixed as well, measure the photodiode current intensity. Remember, one must know the photodiode spectral sensitivity and the actual LED emission wavelength in order to determine the LED irradiance. On our experiments we used three different LED lamps, a violet one emitting at 407 nm (26,71W/m^2) and two UV LEDs at 365 nm, (32,18 W/m^2) and 355 nm (16,40 W/m^2) respectively.

A group of LED lamps is mounted on both sides of the systems with nitrosyl complexes in order to optimize the light exposure; the light up time is controlled by the operator using the constant current power on switch. A constant current power supply drives the LEDs and determines a high stability (< 0.07%) of the LED output light.

ISSN: 2321-9653; IC Value: 45.98; SJ Impact Factor: 7.538
Volume 10 Issue I Jan 2022- Available at www.ijraset.com



Graphic representation 1. The proposed photoreactor top (A) and side (B) views. On the top view (1) indicates the glass reactor, (2) an optional quartz cuvette, (3) the IRpalettes support, (4) the LED lamp mechanical support and (5) the LED lamp itself. (6) a dashed line drawing represents an optional LED lamp for quartz cuvette illumination.



Side View Front View

Graphic representation 2. The LED lamp mounted with nine 5 mm LEDs in its aluminum housing side and front views.

D. FT-IR experiments

Were performed on a BOMEM-MB-102, 4000-400 cm^{-1} range. Freshly prepared KBr pellets with nitrosyl metal complexes alone and mixture scavenger NO. Aiming at an additional control in the release of nitric oxide, as well as biological applications, these complexes were immobilized on organically modified silicates via a sol–gel process, starting with silane tetroxide and 3–glycidoxypropyltrimethoxysilane, these results aren´t reported this paper. The sample was irradiated at room temperature (24 ^{o}C) and recorded the spectrum IR. Spectral resolution was 4 cm^{-1}.

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III. RESULTS AND DISCUSSION

Figure 1 illustrates the change in the vibrational spectrum curve to the *cis*-$[Ru(NO)(NO_2)(phen)_2](PF_6)_2$ complex. The KBr pellets of *cis*-$[Ru(NO)(NO_2)(phen)_2](PF_6)_2$ show a signal at 1945 cm^{-1} and shoulder at 1938 cm^{-1}. Irradiation at 407 nm for 30 s, occurred a decrease in the intensity of absorbance at 1945 cm^{-1} and appearance a band at 1927 cm^{-1} and two broad bands in 1882 cm^{-1} and 1868 cm^{-1} with isosbestic point at 1935 cm^{-1}.

There is a similar change in the complex analogous to bipyridine. A decrease in band absorbance in 1945 cm^{-1}, assigned to stretch N-O with NO^+ character, with a time interval from the first to the second spectrum of 20 seconds, and the appearance of a band in 1926 cm^{-1}. In addition, there is the appearance of a low intensity band in 1882 and 1815 cm^{-1}, the latter less resolved. Signal changes in the range of 1600 cm^{-1} to 1200 cm^{-1} were also verified, which covers vibrational modes related to the NO_2^- centered stretch.

Based on isomerization reactions of the -$[Ru(NO)(NO_2)(bpy)_2](PF_6)_2$ in KBr, studied by KOVALEVSKY et al. [14], which irradiated the samples at 485 nm as use of Ar^+ CW Laser, and in this work with LED light with radiation of 407 nm , the signal in 1882 cm^{-1} attributed to the N-O stretch in NO^+, to the isomer in which NO_2^- is as axial ligand coordinated by oxygen atom and the NO^+ coordinated to the Ru, through the "N" atom, to the complexes with *cis*-$[Ru(NO)(ONO)(X\text{-}Y)_2](PF_6)_2$, for X-Y=phen and bpy, respectively (structure A in scheme 1). In addition, the signals in 1868 and 1871 cm^{-1}, assigned to the N-O stretch in NO^+, in the isomer in which NO_2^-, is also axial ligand and coordinates through the atom of "N" and NO through the "O" in the complex *cis*$[Ru(ON)(NO_2)(X\text{-}Y)_2](PF_6)_2$, for X-Y=phen and bpy, respectively **(B)**. There are also signs in 1926 and 1927 cm^{-1}, isomer **C**, in which no with NO^+ is the axial ligand and coordinates to the ruthenium through the atom of "N" and NO_2^- through the oxygen atom.

Scheme 1: Structures of linkage isomers of *cis*-$[Ru(NO)(NO_2)(phen)_2]^{2+}$

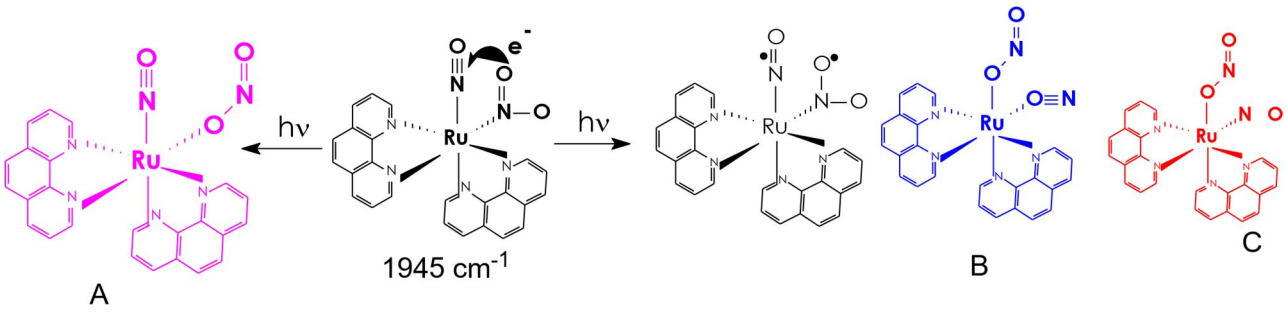


Table 1: Data of υN-O (cm^{-1}) in different intermediates, obtained after irradiation with 407 nm LED light in *cis*-$[Ru(NO)(NO_2)(X\text{-}Y)_2]$ $(PF_6)_2$, in KBr.

X-Y	Isomer	NO^+
Bpy	(A)	1882 (1892*)
	(B)	1815*
	(C)	1926 (1928*)
Phen	(A)	1882
	(B)	1868
	(C)	1927

* [14]

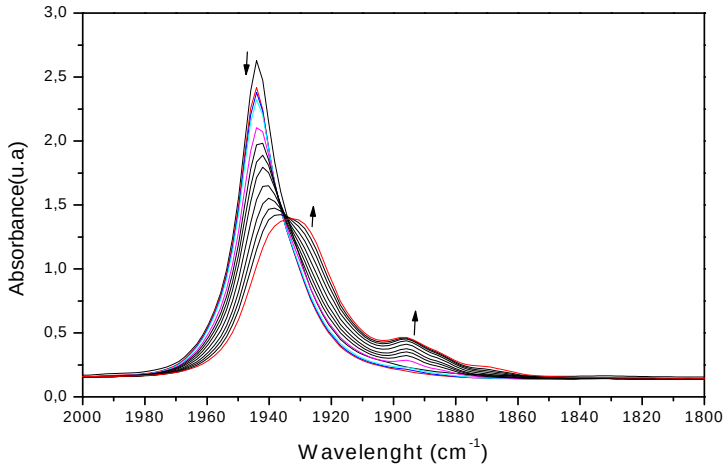


Figure 1- Alteration of the vibrational spectrum for the complex *cis*[Ru(NO)(NO_2)(phen)$_2$](PF_6)$_2$, in KBr, with successive irradiations at. LED of 407 nm.

A. Measured at 30 s Intervals.

Figure 2 shows the spectrum alteration in the infrared region when a KBr pellets containing the *cis*-[Ru(NO)(NO_2)(phen)$_2$](PF_6)$_2$ and [Ru(HEDTA)(OH_2)]complexes (HEDTA= ethylenediaminotetratic acid), a NO scavenger. These experiments were conducted to evaluate the potential of nitrosyl complex *cis*-[Ru(NO)(NO_2)(X-Y)$_2$]$^{2+}$ act as a NO donor.

When comparing figures 1 and 2 a different change can be ratified. It verifies the appearance of a shoulder in the region of 1928 cm^{-1} in Figure 2, which indicates that isomer **C** does not have a favored formation as occurred when the complex is irradiated in the absence of the NO scavenger. It also checks the highlight of a signal in 1890 cm^{-1}, a band with relatively high intensity. Based on the results it can be proposed that the light source used results, in addition to the isomerization reaction, a NO photorelease, which indicates that NO^+ in *cis*-[Ru(NO)(NO_2)(phen)$_2$](PF_6)$_2$ can be reduced and released.

The NO released reacts with [Ru(HEDTA)(OH_2)] and results in [Ru(NO)(HEDTA)(OH_2)], which according to the literature has N-O in NO $^+$ at 1846 cm^{-1} [28]. The signal at 1890 cm^{-1}, illustrated in figure 2, depending on the experimental condition, can be assigned to the formation of the nitrosyl complex from the reaction between released NO and the nitric oxide trap. For the *cis*-[Ru(NO)(NO_2)(bpy)$_2$]](PF_6)$_2$ complex, the experiments conducted in the presence of the capturer, the photoliberation of NO seems to be less favored than in the complex containing phenanthroline. This conclusion is based on changing the vibrational mode N-O^+ in the original complex to energy consistent with the formation of the **C** isomer.

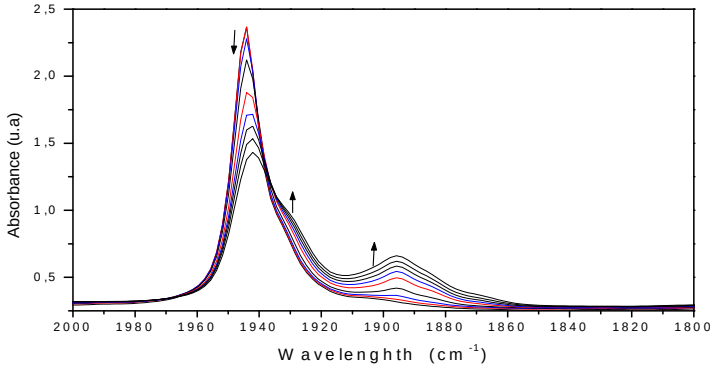


Figure 2- Alteration of the vibrational spectrum for *cis*- Ru(NO)(NO_2)(phen)$_2$](PF_6)$_2$ and [Ru(Hedta)(OH_2)]complex in KBr, with successive irradiations. LED source: Major radiation at 407 nm.

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The support of isomerization in complexes containing NO^+ and NO_2^- coordinated to ruthenium *cis*-[Ru(NO)(NO_2)(phen)2](PF_6)$_2$, was ratified when evaluating the alteration of the NO stretch band with nitrosonium character to the *cis*[Ru(NO)(Cl)(phen)$_2$](PF_6)$_2$ complex. Successive spectra indicated a decrease in band absorbance positioned in 1930 cm^{-1}. This alteration was attributed to the photorelease of NO stimulated by LED light, since the experiments conducted in the presence of the NO scavenger [Ru(Hedta)(OH_2)] indicated: i- Decrease of absorbance in 1930 cm^{-1}. ii- Formation of a band at 2340 cm^{-1} and shoulder in 1890 cm^{-1}.

Considering items i and ii, it can be affirmed that photo-deliberate NO can react with the NO scavenger through an oxirreduction reaction, in addition to a Lewis acid-base reaction, parallel to a redox reaction. These statements are sustained since: a- with the oxidation of NO results in the NO^+ encapsulated in KBr pellets, whose vibrational mode occurs at 2377 cm^{-1} in the compound (NO)(BF_4) [29, 30] near the band illustrated in Figure 3 at 2340 cm^{-1}. Parallel oxidation NO/NO^+ occur the reduction [$Ru^{III/II}$(Hedta)(OH_2)]. b- NO can coordinate to the [Ru^{III}(Hedta)(OH_2)] replacing the water molecule and in parallel the same can be oxidized to NO^+ and Ru(III) is reduced to Ru(II) and thus forms the product [Ru(NO)(Hedta)], therefore the fragment {Ru-NO}^{6}.

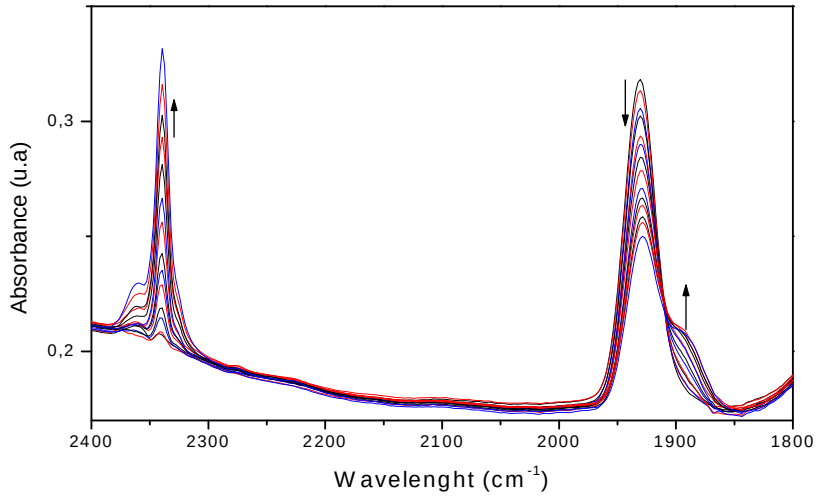


Figure 3- Change of the vibrational spectrum to the complex *cis*[Ru(NO)(Cl)(phen)$_2$](PF_6)$_2$ and [Ru(Hedta)(OH_2)] in KBr, with successive irradiations. LED Source: Majority radiation at 407 nm

In our research group, the formulation complexes *cis*-[Ru(NO)(dye)(XY)](PF_6)$_4$, in which dye are methylene blue (am) and new methylene blue (nam) and XY are the bases of Lewis bipyridine and phenantroline [27]. In this work, the potential of these complexes was evaluated to act as a NO donor when submitted to photochemical stimulus with light source provided for LED. When the complex is supported on KBr pellets and irradiated, with intervals of 60 seconds a decrease in absorbance related to NO^+ stretch (1944 cm^{-1}) in the initial complex and formation bands in 1928 cm^{-1} and 1895 cm^{-1} (Figure 4). In analogy to studies carried out by KOVALEVSKY et al. [14], for the complex *cis*-[Ru(NO)(NO_2)(bpy)$_2$]$^{2+}$, is due to isomerization reactions, whose proposals for formed products require more accurate studies. On the other hand, the experiments conducted in the presence of [Ru(Hedta)(OH_2)] in the same way as it was reported for the *cis*-[Ru(NO)(Cl)(phen)$_2$]](PF_6)$_2$ complex, the alteration of spectra in the region of 2000-1800 cm^{-1}, are consistent with the oxirreduction reaction between NO and the capturer, seen the band formation of NO^+ encapsulated in the KBr pellets at 2340 cm^{-1} [28, 29] as well as an Lewis acid-base parallels the redox reaction leading to the product [Ru(NO)(Hedta)], with the stretch of the N-O with NO^+ character in 1890 cm^{-1}. Therefore, it can be affirmed that the *cis*-[RuNO(dye)(X-Y)](PF_6)$_4$ complexes are NO donors when submitted to radiation with light provided by LEDs with majority radiation in 407 (figure 5), 355 and 365 nm

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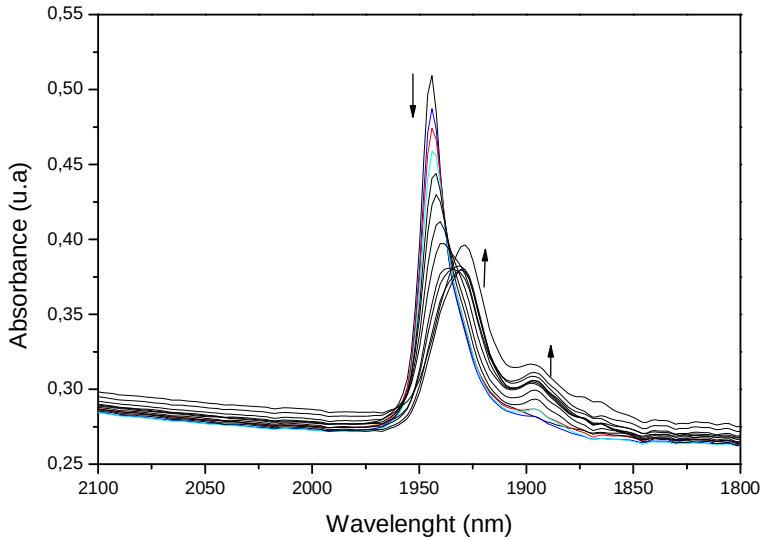


Figure 4- Change of vibrational spectrum to the *cis*-[Ru(NO)(am)$(phen)_2$]$(PF_6)_2$ complex supported in KBr, with successive irradiations. LED Source: Majority radiation at 407 nm

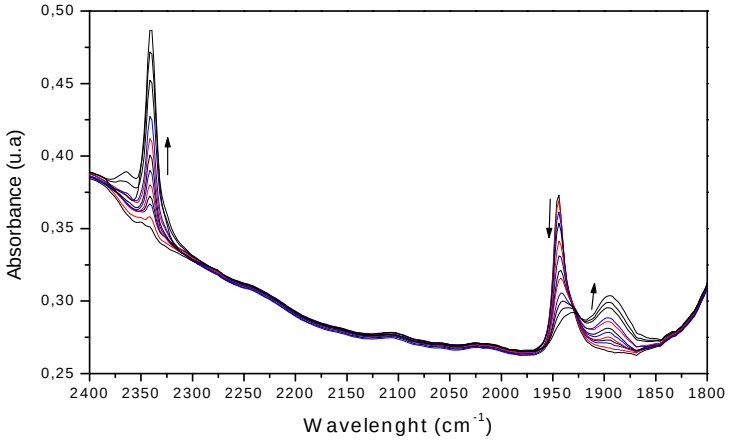


Figure 5- Change of vibrational spectrum to the *cis*-[Ru(NO)(am)$(phen)_2$]$(PF_6)_2$ and [Ru(Hedta)(OH_2)] complex in KBr pellett, with successive irradiations. LED
Source: Majority radiation at 407 nm

One of the nitrosyl complex of therapeutic application is sodium nitroprussiate $Na_2[Fe(CN)_5(NO)]$, which is used in an emergency situation, thus evaluated in this work the behavior when it was submitted to radiation provided of the LED. The results with the use of LED with majority wavelength in 407 nm illustrate a decrease in the intensity of the band related to the N-O of NO^+ and appearance of band in the region of uncoordinated $NO^{+,}$ which is indicative that the photo-deliberate and oxidized NO. The irradiation experiments of a KBr pellet containing $Na_2[Fe^{II}(CN)_5(NO)]$ and [Ru(Hedta)(OH_2)] (figure 6), being the capturer with excess amount of matter compared to nitrosyl, illustrates a change consistent with the formation of uncoordinated NO^+ and [Ru(NO)(Hedta)].

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The products mentioned were also proposed for the *cis*-[Ru(NO)(Cl)(phen)$_2$]](PF$_6$)$_2$ and *cis*-[Ru(NO)(am)(phen)$_2$](PF$_6$)$_2$ systems as a consequence oxireduction and Lewis- acid-base reactions followed by redox reaction, respectively. It was also verified that successive irradiations of Na$_2$[Fe^{II}(CN)$_5$(NO)] result in CN$^-$ photoliberation (decrease in stretch by 2078 cm^{-1} attributed to the coordinated CN$^-$), a consequence that leads to the use of the drug only in emergency situations. The Figure 6 illustrated the existence of a shoulder in 1790 cm^{-1}, which may be due to the NO probably coordinated to the acid ion of Lewis Fe(II).

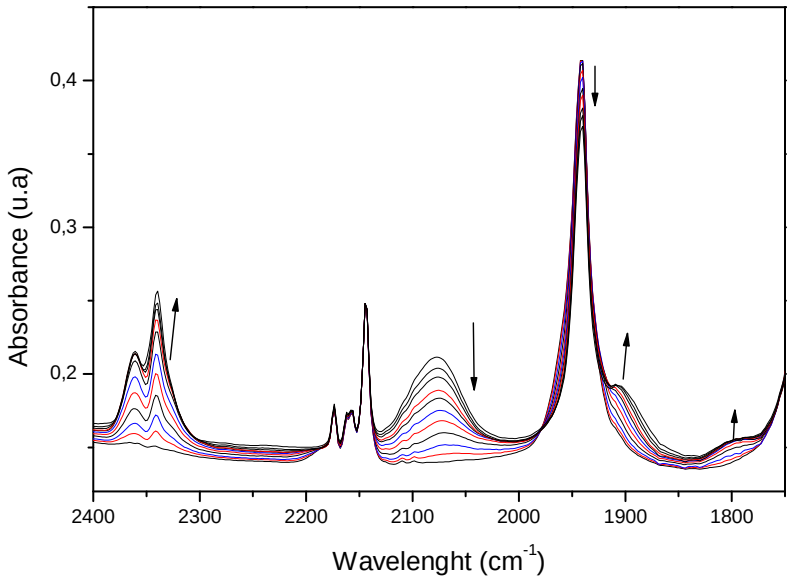


Figure 6- Change of the vibrational spectrum to the Na$_2$[Fe^{II}(CN)$_5$(NO)] and [Ru(Hedta)(OH$_2$)] complex in KBr tablet, with successive irradiations. LED Source: Majority radiation at 407 nm

IV. CONCLUSIONS

The LED system proved to be the feasibility of making use of a reasonably simple system in order to investigate the reactivity of nitrosyl complex. Due to the results obtained, it was possible to ratify the proposal for a photoinduced isomerization of nitrite ligand from (NOO$^-$) to (ONO$^-$) and NO$^+$ for ON$^+$, in the *cis*-[Ru(NO)(NO$_2$)(XY)$_2$](PF$_6$)$_2$ complex, as well as the photoresolution of NO in the complex in which X-Y is phenantroline. For the complex cis-[Ru(NO)(am)(phen)$_2$]$^{2+}$ (am= dye: methylene blue) photoreaction investigation raised the proposal of isomerization and photoliberation reactions of NO, which encourages the investigation of this dye system with potential use in therapy Photodynamic.

For the NO donor metallopharmaceutical, the NPS, with irradiation there was a spectrum change in the region of 2200-1800, which illustrates that there is photoliberation of NO and also CN$^-$. Studies in the presence of NO capturers confirmed that no released in the presence of NO capturer provides an electron transfer reaction with it, in addition to an acid-based reaction from Lewis.

V. ACKNOWLEDGMENTS

The work was supported financially by the Fundação de Amparo a Pesquisa do Estado da Bahia (FAPESB), CAPES and CNPq.

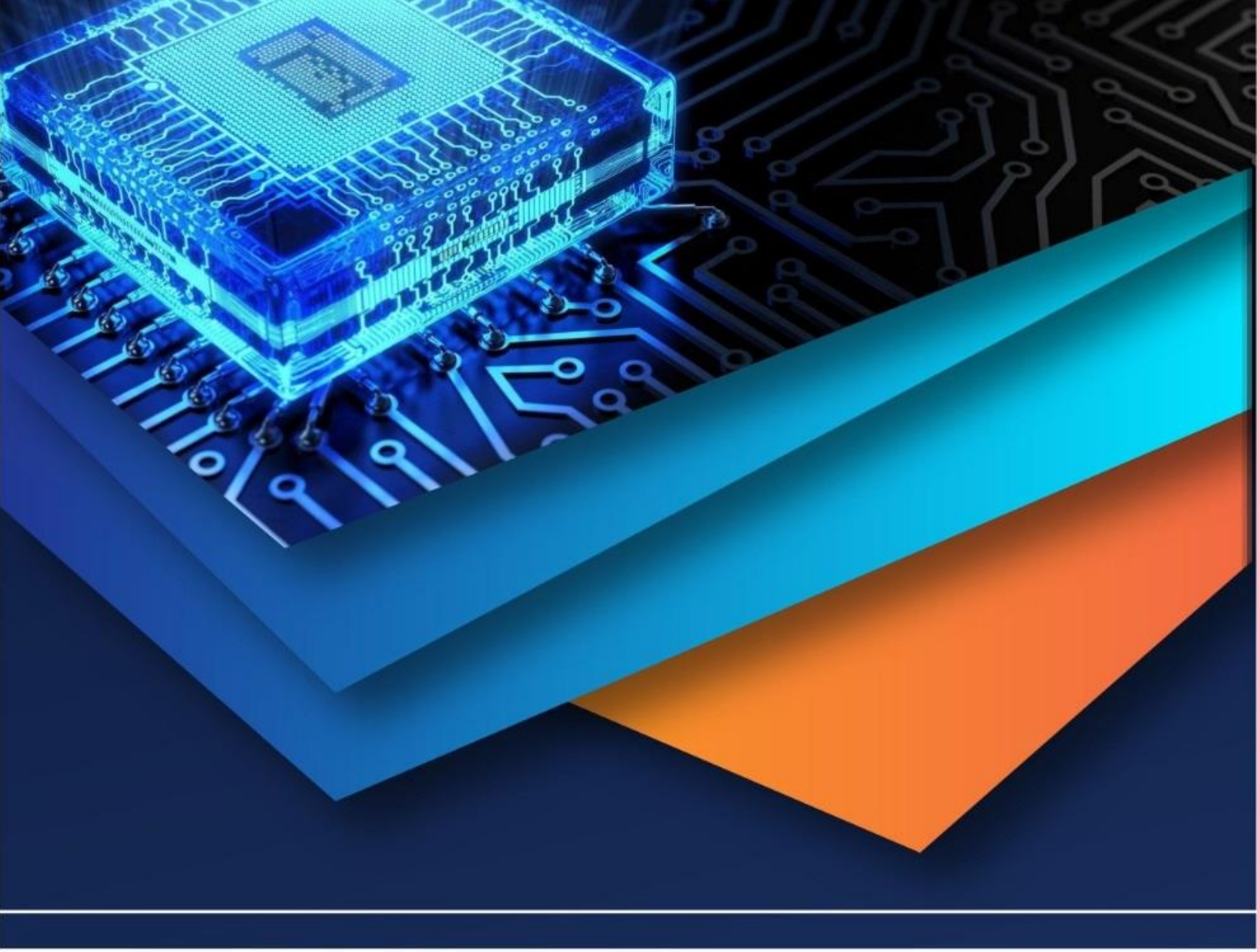
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