Research Journal of Recent Sciences ISSN 2277-2502
Vol. **14(3),** 14-18, July **(2025)** *Res. J. Recent Sci.*

Binding studies of Ru(II) complex with DNA isolated from orange pulp extract

J.R. Jefey, T. Sumitha Celin and G. Allen Gnana Raj
Department of Chemistry and Research Centre, Scott Christian College (Autonomous) Nagercoil (Affiliated to Manonmaniam Sundaranar University, Abishekapatti, Tirunelveli, Tamilnadu, India
sumithaezhil77@gmail.com

Available online at: www.isca.in, www.isca.me
Received 17th March 2025, revised 23rd April 2025, accepted 7th June 2025

Abstract

For a long time, researchers have been closely examining how metal complexes interact with DNA in order to create novel compounds or medications for use in medicine. Ru (II) complexes have shown an excellent DNA binding results. These complexes bind to DNA through intercalative mode. The binding of Ru (II) phenanthroline complex $[RuL_3]^{2+}$ (where L=1, 10-phenanthroline) with DNA of guava fruit extract have been studied in aqueous medium by means of absorption and emission spectral techniques. Using the Benesi-Hildebrand equation, the complex's binding constant (Kb) with the DNA extracted from guava fruit was ascertained. The luminophore and the DNA molecule have a hydrophobic ground state interaction. It was found to be $9.506x10^4M^{-1}$. The ligands in the complex and the purity of the DNA determine the binding constant's value.

Keywords: Luminophore, ligands, absorption, Electrostatic interaction, $[Ru(BPDS)_3]^{2+}$ complex.

Introduction

Because of their applications in biology, metal complexes are believed to be essential in treatment. Among the different transition metals, Ru(II) metal coordination complexes are considered as possible building blocks to design suitable functional materials because of their exceptional photophysical and optoelectronic properties, which primarily evolve from their metal to ligand charge transfer (MLCT) excited states[1–3]. The study of transition metal complexes' interactions with DNA has garnered a lot of attention because of their many applications in biotechnology and cancer treatment[4–9]. A common target molecule for anticancer medications is DNA. To learn more about how the drug molecule interacts with DNA, metal complexes that were bound to DNA were investigated Deoxyribonucleic acid is a polymer composed of two polynucleotide chains that coil around each other to create a double helix. The polymer contains the genetic instructions necessary for all known creatures and numerous viruses to develop, function, grow, and reproduce. DNA is a lengthy polymer composed of nucleotides, which are repeating units. DNA has a dynamic structure that can coil into tight loops and other configurations along its length.

Interactions with other molecules are one of the many situations that might cause damage to DNA molecules. This damage may cause a range of pathogenic changes in live organisms because of their potential as novel therapeutic agents and their photochemical properties, which make them potential probes of DNA structure and conformation[10]. Ru(II) bathophenanthroline disulphonate complexes exhibit potent DNA binding capabilities. These complexes bind to DNA using the intercalative technique. This work highlights the interaction of Ru complexes with DNA derived from orange pulp extract in light of the backdrop.

Materials and Methods

Materials: The ligand bathophenanthroline disulphonate, which was purchased from Sigma Aldrich, was utilized without any purification. By reacting $RuCl_3.3H_2O$ with the proper ligands, the complex was created in accordance with the previously reported protocol[11]. Binding studies were carried out using double distilled water. We bought orange, detergent, ethanol, sodium chloride, and liquid soap from local supermarkets. A 95% concentration of ethanol was used for the DNA isolation.

Synthesis of $[Ru(BPDS)_3]Cl_2$ complex: 1g of $RuCl_3.H_2O$ is dissolved in 50mL water and 50 mL ethanol is added. This mixture was refluxed in a hot water bath for 4-5 hrs and a blue solution is got. 5mL of this solution was transferred into a degassed solution of disodium phenanthroline di-yl-4,7-diphenyl sulphonate (1g) in 20 mL water and is refluxed under nitrogen for 12 hrs. The resultant solution was filtered and evaporated to get orange red crystal. We bought orange, detergent, ethanol, sodium chloride, and liquid soap from local supermarkets. A 95% concentration of ethanol was used for the DNA isolation.

Extraction of DNA fragments: Chopped orange was used in tiny bits. In a beaker with roughly 20 milliliters of water, two table spoons of regular salt were added. There were two tablespoons of dish soap added. The detergent solution, minced orange, and table salt were all mashed together.

The mixture is then poured into a fresh beaker. A tea strainer was then used to spin the liquid over a flame before filtering it out into a fresh beaker. Five minutes were used to set it aside. Ethanol (10mL) was added ice cold. After a short time, white strands of DNA were visible. Condensing in the ethanol layer, the DNA separates out. A glass rod is used to spoole on the generated DNA.

Equipments: All the spectral measurements were carried out using Systronics-2203 UV-Visible double beam spectrophotometer. Emission measurements were done using JASCO-FP 8200 spectrofluorometer. All spectral measurements were taken at room temperature.

Determination of Binding constants: The $[Ru(BPDS)_3]$ association constants (K_a^{abs}) Benesi-Hildebrand technique (1) was used to calculate binding of the complexes with DNA isolated from orange pulp extract in homogenous medium.

$$\frac{1}{\Delta A} = \frac{1}{K_a^{abs}}\Delta\varepsilon[H] + \frac{1}{\Delta\varepsilon}[Q] \quad (1)$$

Here: [H] stands for host (sensitizer) concentration, [Q] for guest (quencher) concentration, and /A for the change in absorbance of [H] upon the addition of [Q]. The molar extinction coefficient of the free [H] and [H]-[Q] complexes differs. The 1:1 complex formation is supported by the plot of 1/A values as a function of 1/[Q] values for each of the guest molecules that were studied. The ratio of the Y-intercept to the straight line's slope yields the association constant[13].

Results and Discussion

The structure of complex is shown in Figure-1. $[Ru(BPDS)_3]^{2+}$ shows an absorption maximum at 445 nm and an emission maximum at 600 nm in water. The emission maximum of Ru(II) complexes originates from the dπ-π* ^{3}MLCT transition. The lowest excited state of $[Ru(phen)_3]^{2+}$ is a triplet metal to ligand charge transfer state ^{3}MLCT excited state. The binding takes place in the LC and in the MLCT absorption maximum of the complex in the ground state.

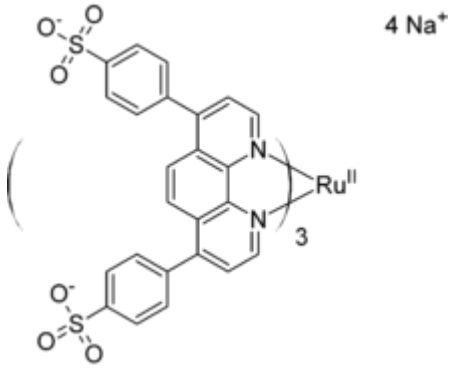

Figure-1: Structure of $Ru(BPDS)_3]^{2+}$ complex.

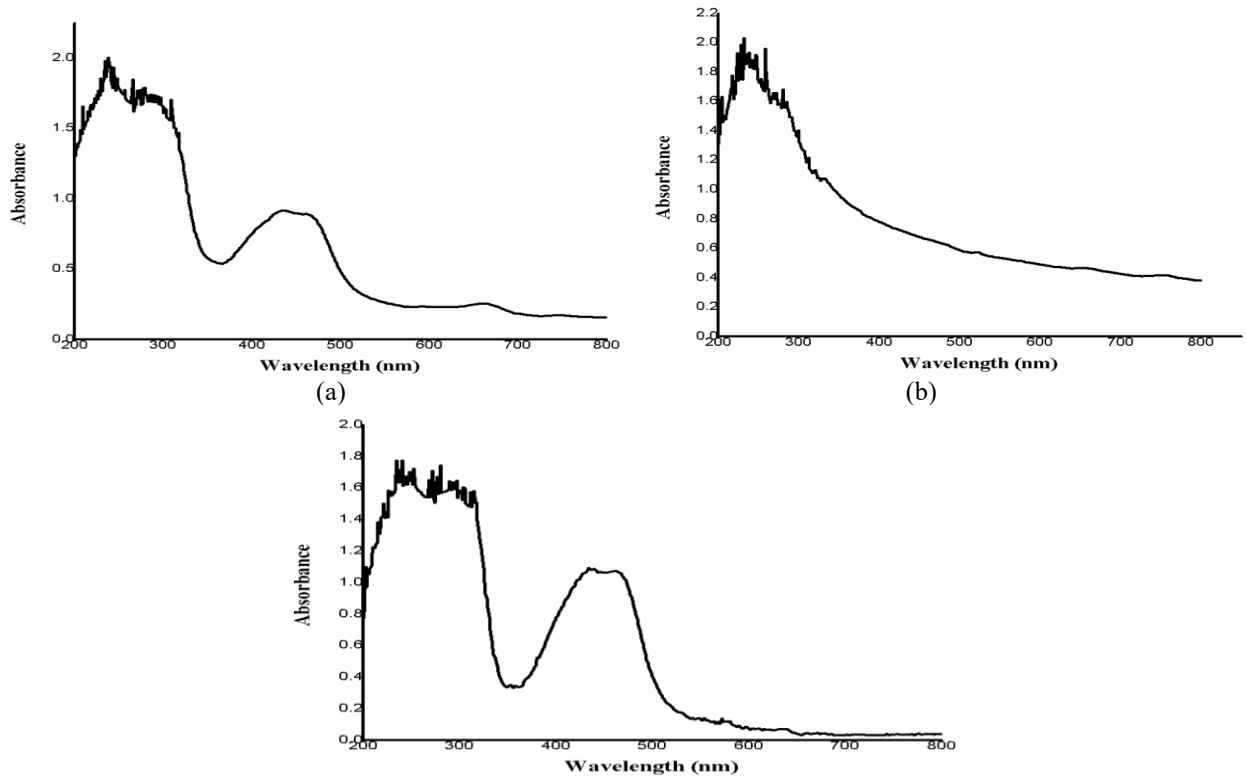

Figure-2: Absorption spectrum of (a) $[Ru(BPDS)_3]^{2+}$ complex (b) DNA of Orange pulp extract (c) $[Ru(BPDS)_3]^{2+}$ complex with Orange pulp DNA extract.

The $[Ru(BPDS)_3]^{2+}$ complex absorption spectrum, with orange pulp DNA extract, is displayed in the Figure-2. It exhibits a shoulder peak at 445 nm and in the region at 279 nm there is high energy absorption.

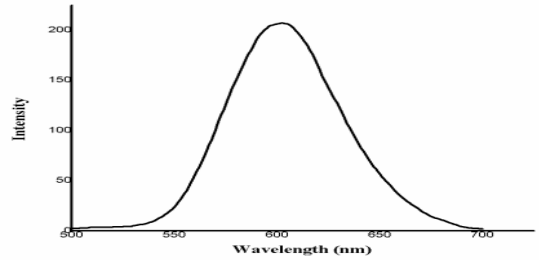

Figure-3: Emission spectra of $[Ru(BPDS)_3]^{2+}$ with orange pulp DNA extract in aqueous media.

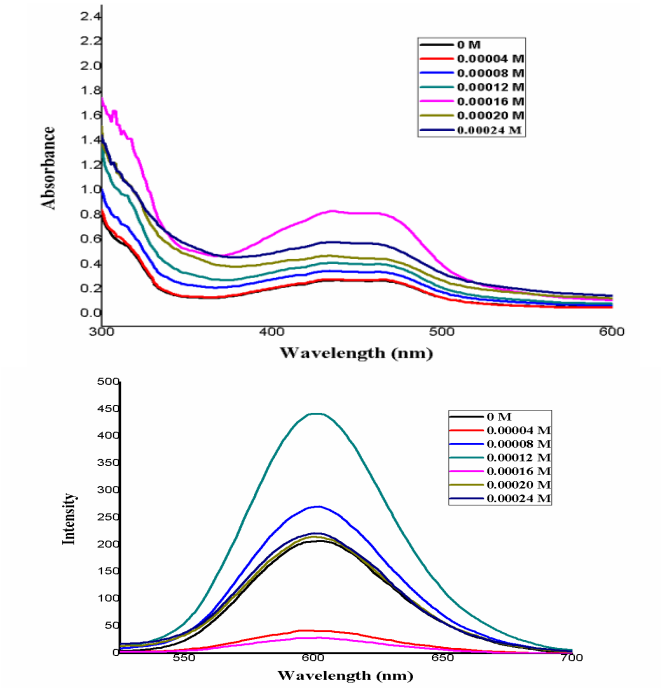

Figure-4: (a) Absorption spectra (b) Emission spectra of $Ru(BPDS)_3]^{2+}$ with incremental concentration of orange fruit DNA extract in aqueous media.

These data obtained from absorption and emission spectrum data are used to calculate the DNA fruit's binding with different metal complexes. In order to maintain a total volume of 5 mL for the DNA metal complex solution, the concentration of the metal complex was maintained constant while the concentration of DNA extracted from fruit was changed. Measurements of absorption and emission were made for different amounts of complex DNA. The change in absorbance was computed for the absorption measurements. Using the emission spectrum data, the changes in emission intensity was also computed. The binding constant for the DNA-metal interaction was calculated using these calculations. The Benesi-Hildebrand plot is used for this.

Both the absorption and emission measurements are used to display the metal complex's Benesi-Hildebrand plot. The binding constants for these plot were calculated by taking the ratio of the intercept and the slope. Based on the combined absorbance and luminescence intensity released by the metal complex in the presence of varying DNA quantities, the association constant (Ka) for the production of adducts between DNA and photoexcited Ru(II)-complexes was calculated. The values of the association constant K_a (M^{-1}) for DNA with $[Ru(NN)_3]^{2+}$ complexes in aqueous medium fall between 10^4 and $10^5 M^{-1}$.

Figure-4 shows the $[Ru(BPDS)_3]^{2+}$ complex's absorption and emission maximum in an aqueous medium after orange pulp extract DNA(G) is gradually added. The change in the absorbance of the DNA with the gradual addition of the complex indicates the complex's binding affinity. The complex's ground-state interactions with the DNA are either π-stacking or hydrophobic. Based on the Benesi-Hildebrand plot, the K_b of the $[Ru(BPDS)_3]^{2+}$ complex with the isolated orange pulp DNA is $5.33x10^4$ M^{-1} at the LC area and $9.506x10^4$ M^{-1} at the MLCT region, respectively. The outcome shows that the MLCT region of the DNA binds more firmly than the LC region.

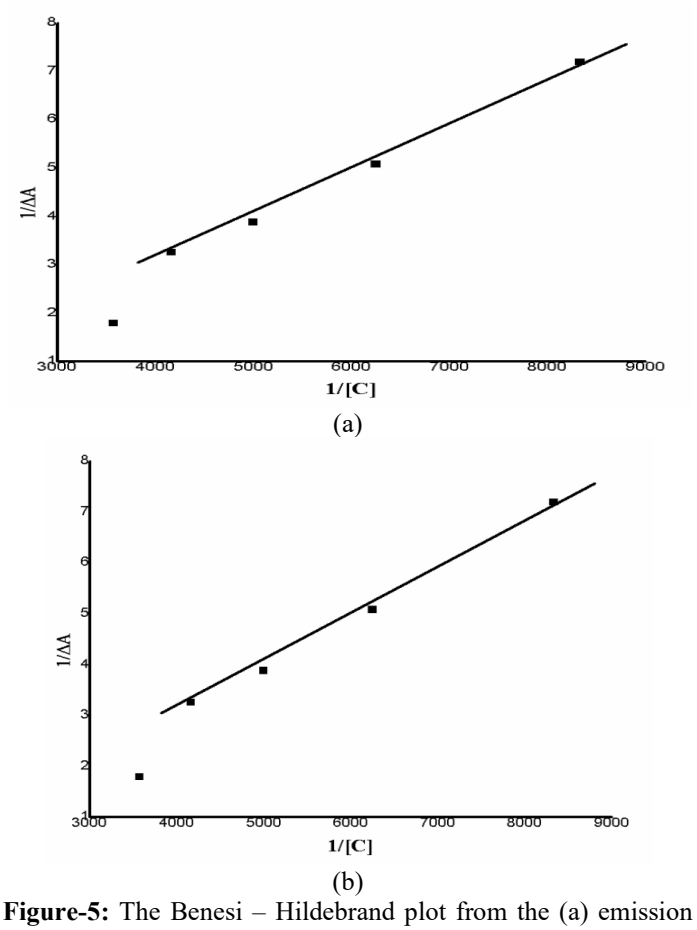

Figure-5: The Benesi – Hildebrand plot from the (a) emission spectral data (b) absorption spectral data of $[Ru(BPDS)_3]^{2+}$ with orange pulp DNA extract in aqueous media.

Hydrophobic or stacking interactions characterise the ground state interaction between isolated DNA and the bathophenanthroline disulphonate rings of the luminophore. The interaction between stacking and binding makes binding stronger. The results reveal that the complex binds with the orange pulp DNA strongly. The binding of the complex depends on the ligands present in the complex as well as the purity of the DNA. The complex interacts with the DNA both by electrostatic as well as intercalative mode of binding. This type of binding leads to the breakage of the DNA double helical structure. The hydrogen bonding formation between the DNA base pairs and also the Van der Waals interactions between the complex and the base pairs of the DNA leads to strong binding[14-18]. The result interprets the strong binding of the complex with the DNA isolated from orange pulp extract.

Conclusion

The binding of $[Ru(BPDS)_3]^{2+}$ complex with the DNA extracted from orange pulp has been investigated using both absorption and emission spectral techniques. It has been investigated how the DNA extract bind to the Ruthenium (II) complexes. The evaluation of binding constants were described in detail. According to our current research, the DNA extract has a strong affinity for $[Ru(BPDS)_3]^{2+}$ complex. It has the highest binding constant with the value of 9.506×10^4. This higher binding constant shows good interaction, which is crucial as it suggests that the complexes can effectively bind to DNA, which is a key factor in their potential therapeutic applications.

Acknowledgement

The authors thank the management for providing the DST-FIST lab facilities and the Chemistry Department of Scott Christian College (Autonomous), Nagercoil for carrying out the project work.

References

1. Bar, M., Deb, S., Paul, A. & Baitalik, S. (2018). Stimuli-responsive luminescent bis-tridentate Ru(II) complexes toward the design of functional materials. *Inorg. Chem*., 57, 12010-12024.

2. Balzani, V., Juris, A., Venturi, M., Campagna, S., & Serroni, S. (1996). Luminescent and redox-active polynuclear transition metal complexes. *Chem. Rev*., 96, 759-834.

3. Sauvage, J.P., Collin, J.P.J., Chambron, C., Guillerez, S., Coudret, C., Balzani, V., Barigelletti, F., Cola, D.L., & Flamigni, L. (1994). Ruthenium(II) and Osmium(II) bis (terpyridine) complexes in covalently-linked multicomponent systems: Synthesis, electrochemical behaviour, absorption spectra, and photochemical and photophysical properties. *Chem. Rev*., 94, 993-1019.

4. Gomathi, R., Ramu, A., & Murugan, A. (2013). Synthesis, spectral characterization of N-benzyl isatinschiff base Cu(II), Co(II) and Ni(II) complexes and their effect on cancer cell lines. *International Journal of Innovative Research in Science, Engineering and Technology*, 10, 5156 - 5166.

5. Gomathi, R., and Ramu, A. (2013). Synthesis, DNA binding, cleavage, antibacterial and cytotoxic activity of Novel Schiff base Co(II) Complexes of substituted isatin. *International Journal of Advanced Research*, 1(8), 556–567.

6. Rosenberg, B., Van Camp, L. and Krigas, T. (1965). Inhibiton of cell division in *Escherichia coli* by electrolysis products from a platinum electrode. *Nature,* 205, 698-699.

7. Barton, J. K. and Lolis E. (1985). Chiral discrimination in the covalent binding of bis(phenanthroline) dichloro ruthenium(II) to B-DNA. *Journal of the American Chemical Society,* 107(3), 708-709. https://doi.org/10.1021/ja00289a035

8. Guo, Z., and Sadler, P. J. (2000). Medicinal Inorganic Chemistry. *Advances in Inorganic Chemistry,* 49, 183-184. https://doi.org/10.1016/S0898-8838(08)60271-8.

9. Erkkila, K. E., Odom, D. T. & Barton, J. K. (1999). Recognition and reaction of metallointercalators with DNA. *Chemical Reviews,* 99(9), 2777-2796. https://doi.org/10.1021/cr980434

10. Lippert, B. (2000). Multiplicity of metal ion binding patterns to nucleo bases. *Coordination Chemistry Reviews,* 200, 487-516.

11. Li,C., Liu, S. L., Guo, L. H., and Chen, D. P. (2005). A new chemically amplified electrochemical system for DNA detection in solution Electrochem. *Commuications Chemistry,* 7(1), 23-28.

12. Swarnalatha, K., Rajkumar, E., Rajagopal, S., Ramaraj, R., Banu, I. S. & Ramamurthy, P. (2011). Proton coupled electron transfer reaction of phenols with excited state ruthenium (II)–polypyridyl complexes. *Journal of Physical Organic Chemistry*, 24(1), 14-21.

13. Sumitha Celin, T. & Allen Gnana Raj, G. (2020). Luminescence quenching of tris(4,4′-dimethyl-2,2′-bipyridyl ruthenium (II) complex with quinones in aprotic polar medium. *Indian Journal of Chemistry*, 59A, 923-928.

14. Nordell, K. J., Jackelen, A. L., Condren, S. M., Lisensky, G. C., & Ellis, A. B. (1999). Liver and Onions: DNA Extraction from Animals and Plant Tissues. *Journal of Chemical Education,*76(3), 400A,

15. Saha, B. & Stanbury, D. M. (2000). Thermal and Photochemical Reduction of Aqueous Chlorine by Ruthenium (II) Polypyridyl Complexes. *Inorganic Chemistry*, 39(6), 1294-1300.

16. Abisha, M., Sumitha Celin, T. and Allen Gnana Raj, G. (2022). A study of binding of DNA extracted from onion

with Ruthenium polpyridyl complexes. *Research Journal of Chemical Sciences*, 12(3), 20-24.

17. Jefey J.R, Sumitha Celin T (2023). Binding Studies of Ru(II) Bathophenanthroline Complex With DNA Isolated From guava fruit Extract. Proceedings of National Conference on Biomaterials, Scott Christian College (Autonomous), Nagercoil, India, 21st April. pp 15-19.

18. Sumitha Celin T (2019). Photophysics and Photochemistry of metal polypyridyl complexes. Doctoral Thesis, Manonmaniam Sundaranar University, Tamilnadu, India, pp 1- 221.