Geliş Tarihi/Received Date: 2017-07-17 Kabul Tarihi/Accepted Date: 2017-09-25

DNA-binding and spectroscopic studies of cobalt (II) compound containing 2, 10, 16, 24-tetrakis 4-(2-phenoxy-1, 3-dioxo-2, 3-dihydro-1H-inden-2-yloxy) phthalonitrile

Ali Arslantas[1*], M. Salih Agirtas[2]
[1] Biomedical Engineering Department, School of Engineering, Karabuk University,78050, Karabuk, Turkey
[2] Chemistry Department, Faculty of Science, Yüzüncü Yıl University, 65080, Van, Turkey

Abstract

In this study, binding activities of the previously synthesized cobalt (II) compound (**PcCo**) with calf thymus-DNA containing 2, 10, 16, 24-tetrakis 4-(2-phenoxy-1, 3-dioxo-2, 3-dihydro-1H-inden-2-yloxy) phthalonitrile were investigated to determine binding activities by using UV-Vis absorption, fluorescence titration methods. Absorption titration spectra of **PcCo** in DMF give peaks at about 670 nm for Q-band absorption and about 340 nm for B-band absorption in the absence of calf thymus-DNA. Emission intensities of **PcCo** in presence of calf thymus-DNA, **PcCo** gives emission at about 545 nm. Thermal denaturation profile study was systematically studied by UV/Vis spectroscopic technique and melting point temperature was found around 7.87 ^{o}C for Co (II) phthalocyanine compound**.** The reduction oxidation behavior of **PcCo** with calf thymus-DNA was investigated using cyclic voltammetry (CV). Cathodic peak potential and anodic peak potential for PcCo were recorded to be 0.84, -0.36 V for EPc and 0.88 V, 0.15 V, and -0.69 V EPa. The cyclic voltammetric results show one half-reversible reduction oxidation wave. The shift in Epc and Epa potentials indicates that **PcCo** shows strong interaction with calf thymus-DNA. Further to understand binding activities of **PcCo** with calf thymus-DNA was studied by using electrophoresis technique. Electrophoresis findings showed that there is decrease in the intensity of CT-DNA bands. All the findings show that **PcCo** interacts strongly with DNA molecule via an intercaltion binding.

Keywords: Phthalocyanines, DNA-binding, UV/Vis spectroscopy, gel electrophoresis, cyclic voltammetry.

2, 10, 16, 24-Tetrakis 4- (2-fenoksi-l, 3-diokso-2,3-dihidro-lH-inden-2-iloksi) ftalonitril içeren kobalt (II) bileşiğinin DNA bağlanmasının spektroskopik yöntemler ile araştırılması

Öz

2,10, 16, 24-tetrakis 4- (2-fenoksi-1,3-diokso-2,3-dihidro-lH-inden-2-iloksi) ftalonitril içeren önceden sentezlenmiş kobalt (II) bileşiğinin (**PcCo**) CT-DNA ile bağlanma özellikleri UV/Vis absorpsiyon, floresan titrasyonu yöntemleri kullanılarak bağlanma özelliklerini belirlemek için araştırıldı. DMF çözücü ortamında ve DNA'nın yokluğunda, PcCo'nun absorpsiyon titrasyon spektrumu, Q-band için yaklaşık 670 nm'de pik ve B -bandı için 340 nm de pik verir. Calf thymus-DNA, PcCo varlığında PcCo'nun emisyon yoğunluğu yaklaşık 545 nm'de emisyona neden olur. Termal denatürasyon profili çalışması UV-Vis spektroskopik tekniği ile sistematik olarak incelenmiş ve Co (II) ftalosiyanin bileşiği için erime noktası sıcaklık değeri 7.87 ^{o}C civarında bulunmuştur. CT-DNA yokluğunda ve varlığında **PcCo**'in redüksiyon oksidasyon davranışı, dönüşümlü voltametri (CV) ile incelendi. PcCo için katodik pik ve anodik pik potansiyelleri, EPc için 0.84, -0.36 V ve 0.88 V, 0.15 V ve EPa için -0.69 V olarak kaydedildi. Dönüşümlü (cyclic) voltametri sonuçlar, bir yarı-tersinir redaksiyon oksidasyon dalgasını göstermektedir. Epc ve Epa potansiyelindeki değişim, **PcCo**'in CT-DNA'yla güçlü etkileşim gösterdiğini önermektedir. **PcCo**'in CT-DNA ile olan etkileşimi ayrıca jel elektroforez yöntemi kullanılarak araştırılmıştır. Jel elektroforezi sonuçları, CT-DNA bantlarının yoğunluğunda azalma olduğunu gösterdi. Tüm bulgular, **PcCo** ile CT-DNA molekülü arasındaki etkileşimin interkalatif bağlanma türü olduğunu göstermektedir.

Anaktar Kelimeler: DNA, DNA-bağlanma, Ftalosiyaninler, Jel Elektroforez, UV/Vis spektroskopi

*Sorumlu yazar: Biomedical Engineering Department, School of Engineering, Karabuk University,78050, Karabuk, Turkey, E-mail Adres: arsoz33@gmail.com, Phone: +903704187050

DOI: 10.21541/apjes.329065

1. Introduction

Phthalocyanine metal compounds exhibit substantial electrochemical, physical and spectroscopic features because of their strong conjugative π systems [1-4]. Metellophthalocyanine compounds and their relevant analogs are potential functional substances for usage as dyes and pigments [5-8], chemical sensors [9-11] and photosensititizers for photodynamic therapy for cancer [12, 13]. Many studies conducted to change these kind of macrocyclic metal compounds with the objective of soothing their features and improving their efficiency for advanced substances.

Nowadays, photodynamic treatments have gotten great attention as a promising cancer therapy. Cancer is an important health problem which leads to widespread cause of death in the worldwide [14]. In order to cure abnormal cell of cancer, abnormal growth of these cells must be prevented [15]. In this case, DNA molecule plays an significant role in cell reproduction and genetic mutation. Therefore, DNA molecule is a major target to discover and developing of new anticancer curing medicine [16, 17].

Though the most prevalent therapy methods for abnormal cells of cancer are surgical operations, radiotherapy and chemotherapy, but these types of therapy have substantially side effects [18]. Consequently, scientists proceed to search for new techniques for curing of cancer cell for instance photodynamic treatment is base upon production of reactive oxygen breeds underneath irradiation of photosensitizer. The formation of reactive oxygen species exterminate cancer tumours by necrosis process [19]. Phthalocyanine compounds have been used as photosensitizer materials for photodynamic therapy due to eminent single oxygen product, their stability, strong absorption property and low toxicity [17, 20].

Due to the biological significance of phthalocyanine compounds, it is useful to investigate the binding activities of this type fucntional compound with DNA molecule. In this study, we invastigated the binding activities of previously synthesized cobalt (II) phthalocyanine compound (**PcCo**) [21], bearing four 4-(2-phenoxy-1, 3-dioxo-2, 3-dihydro-1H-inden-2-yloxy) phthalonitrile groups by calf thymus (CT)-DNA using UVVVis absorption spectra, fluorescence spectroscopy, cyclic voltammetry, thermal denaturation profile study and viscosity measurements.

2. Experimental

2.1. Chemical reagents

In this study, used chemicals were molecular biology grade and they were used without purification. Calf thymus-DNA and hydroxymethylaminomethane hydrochloride (Tris-HCl) were purchased from Sigma-Aldricht. Sodium chloride was purchased from Merck. DNA-binding studies were conducted in a Tris-HCl buffer solution, 20 mM Tris-HCl, 20 mM NaCl at pH 7.0. the solution of Tris-HCl buffer was prepared by using Milli-Q water. 2, 10, 16, 24-tetrakis (4-(2-phenoxy-1, 3-dioxo-2, 3-dihydro-1H-inden-2-yloxy) phthalonitrile) phthalocyaninato cobalt (II) compound which is synthesized and characterized according to literature procedure [17].

In this paper, all the experiments were conducted by using the buffer solution at pH 7.0. Calf thymus-DNA solution in Tris-HCl buffer indicated UV/Vis absorbance at 260 nm, demonstrating that calf thymus-DNA solution was protein free [22].

2.2. Equipments

UV-Vis absorption studies of DNA-binding of Co (II) phthalocyanine compound **(PcCo)** with calf thymus-DNA were conducted in aquartz cuvette using Agilent Technologies Cary 60 UV-vis spectrometer (Karabuk, MARGEM, Turkey), fluorescence spectroscopy measurements were recorded with Perkin Elmer LS fluorescence spectrometer and cyclic voltammetry (CV) measurements were carried out using Ivisumstat Electrochemical Interface electrochemical analyzer.

Agarose gel electrophoresis experiments were performed with Thermo owl electrophoresis system. Thermal denaturation temperature of DNA was conducted at 260 nm by using Agilent Technologies Cary 60 UV/Vis spectroscopy and viscosity measurement experiments were conducted out by using Ubbelohde viscometer.

2.3. Synthesis

2.3.1. Synthesis of compound

The compound which was synthesized and characterized according to literature procedure by M.S. Agirtas [21].

2.3.2. 2, 10, 16, 24-tetrakis 4-(2-phenoxy-1, 3-dioxo-2, 3-dihydro-1H-inden-2-yloxy) phthalonitrile}phthalocyaninatocobalt (II) (PcCo)

The compound which was synthesized and characterized according to literature procedure by M.S. Agirtas [21].

2.4. Absorption titration studies of DNA-binding

Absorption spectra measurements were conducted with diffentiating calf thymus-DNA concentrations between the range of 1µM, 1.5µM, 2.5µM and 3.5µM and keeping the concentration of Co (II) phthalocyanine compound constant. On measuring of absorption spectra, certain amount of calf thymus-DNA was added to Co (II) phthalocyanine compound solution. Absorption spectra experiment were carried out on successive addition of calf thymus-DNA sample. Absorption titration spectra of the solutions was recorded between 300 and 800 nm [23]. Fluorescence titration were conducted between 550 and 780 nm after excitating. Fluorescence titration was performed by adding small amount of a concentrated calf thymus-DNA to **PcCo** sample at fixed concentration. Before measurements, the sample solution was allowed to reach to equilibrium for a certain period of time [24].

Melting point temperature experiment was performed by recording the change in the absorption of the DNA at 260 at different temperatures to evaluate thermal denaturation temperature of calf thymus-DNA. The absence and presence of **PcCo** in the buffer at pH 7.0, the melting point temperature was measured containing DNA and **PcCo** samples**.** The sample solution was stirred and the temperature was increased gradually from 25 to 95 ºC with a recording of absorbance taken every 5 ºC. Melting temperatures were recorded at 260 nm wavelenght [25] by using Agilent Technologies Cary 60 UV/Vis spectroscopy.

2.5. Cyclic voltammetric(CV) measurements

All the cyclic voltmmetry experiments were recorded at room temperature in a 10 mL electrolic cell using the buffer solution as supporting electrolyte. Cyclic voltametry experiments were conducted by using Ivisumstat Electrochemical Interface Electrochemical analyzer at the following setting. Beginning potential was -1.5 V and end potential was 1.5 V and the rate of scan was 10 mV/s. Glassy carbon working electrode was an Ag/AgCl reference electrode and plantinnum wire was a counter electrode were used in this study [26, 27]. Cyclic voltammogram of 10 mL of **PcCo** was carried out differentiating concentrations of DNA sample.

2. 6. DNA gel electrophoresis studies

The binding experiment of **PcCo** with the DNA was carried out by electrophoresis. The solution containing calf thymus-DNA and different concentrations of **PcCo** in Tris-HCl buffer was incubated for certain time. Gel electrophoresis experiment was carried out at 80 V for 3 h in TBE buffer solution. DNA bands were visualized by using Vilber Lourmat UV lamb. In this experiment, Thermo owl gel electrophoresis system was used.

2.7. Viscosity measurement studies

In this study, viscosity measurements of **PcCo** with DNA were conducted using Ubblohde viscometer which was submerged in a bath maintaining at a constant temperature (30 ºC). The approximate flow time was achieved after experiments were repeated three times. Realtive viscosity of the DNA was calculated by using $\eta_i=(t_i-t_0)/t_0$ [28].

3. Results and Discussions

3.1. the Synthesis and characterization of PcCo

Co (II) pthalocyanine compound (**PcCo**) bearing 4-(2-phenoxy-1, 3-dioxo-2, 3-dihydro-1H-inden-2-yloxy) phthalonitrile substituents on the peripherial positions was synthesized and characterized according to literature procedure [21]. The chemical structure of tetra substituted Co (II) phthalocyanine compound is given in Fig. 1. The characterization of previously synthesized Co (II) phthalocyanine compound was done by NMR, FTIR and UV/Vis spectroscopy analyses and their results are with reference to literature [21**]**.

3.2. DNA binding studies of PcCo

3.2.1. Absorption Spectra and fluorescence spectroscopy studies of PcCo

Phthalocyanine compounds indicate characteristic electronic spectra with two strong absorption spectra in their ground state absorption. First one of these absorption spectra bands, which is known Q band, which is observed at about 600-750 nm in visible region of spectrum because of transitions between molecular orbitals. Second absorption band, which is known as B band, is observed in the ultraviolet region of spectrum at about 300-450 [29]. When a photosensor compound is used for photodynamic therapy, it must not be toxic effect when it absorps strongly in the region 600-800 nm.

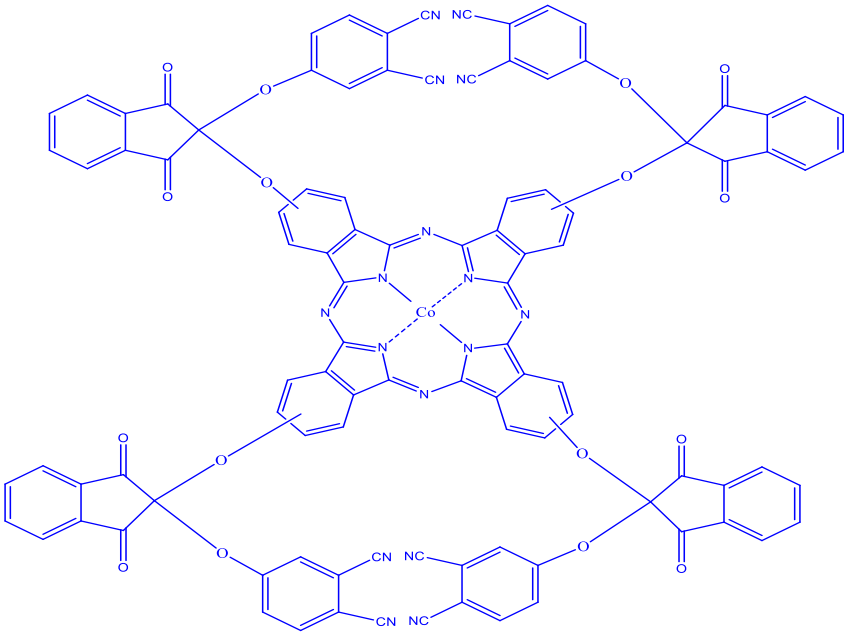

Fig. 1. Chemical structure of cobalt (II) phthalocyanine compound (**PcCo**).

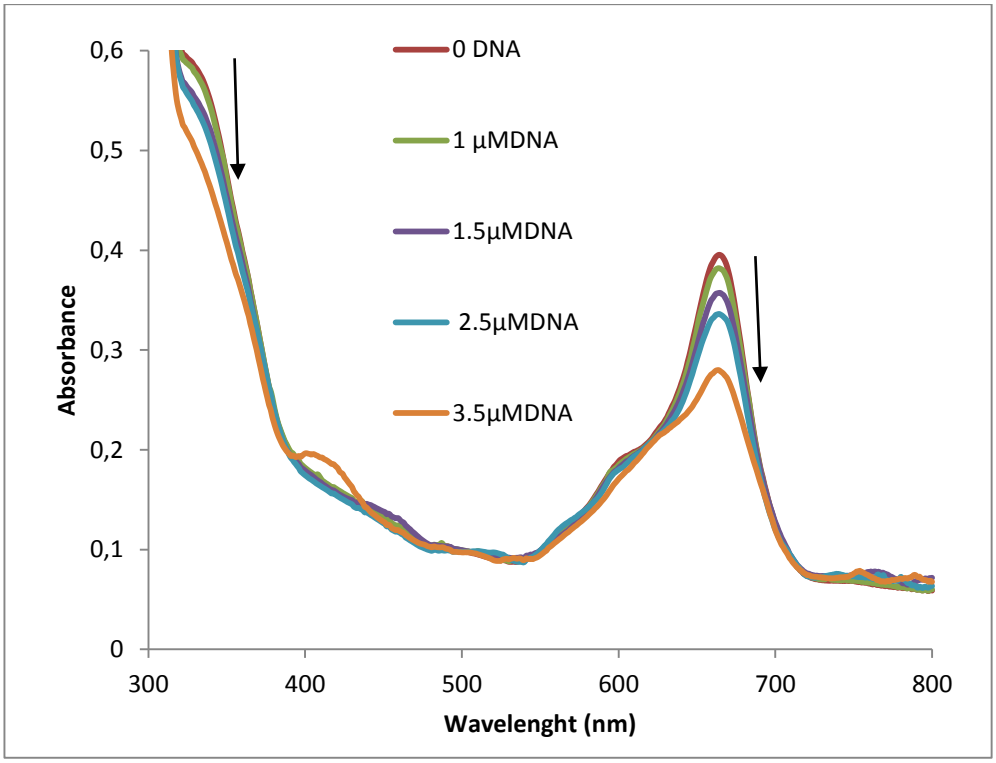

Fig. 2. Absorbance titration spectra of **PcCo** in the buffer in the absence and presense of calf thymus-DNA on addition of calf thymus-DNA from top to bottom. Arrows indicates absorbance change on increasing concentration of calf thymus-DNA from top to bottom.

Absorption spectra were conducted to determine binding activities of Co (II) phthalocyanine compound with DNA molecule. Metallophthalocyanine compounds can react with CT-DNA through non-intercalation and intercalation binding modes. In general, intercalation binding of a small molecule to DNA causes changes in absorbance spectra (hyperchromism or hypochromism) and a red or blue change in wavelength [29] of compounds due to π-packing

interaction between aromatic groups [18] in comperison with DNA electrostatic binding agents cause small changes in absorbances and wavelengths in absorption spectra [16, 30]. An interaction such as intercalation binding mode generally is related to a hypochromic and a red change [29]

In this study, the absorbance titration spectra of **PcCo** were performed to determine binding activities between DNA and **PcCo**. For DNA-binding of **PcCo**, absorption titration experiments were carried out between the range of 300 and 800 nm. Absorption titration spectra of **PcCo** in DMF give peaks at about 670 nm for Q-band absorption and about 340 nm for B-band absorption in the absence of calf thymus-DNA. Absorption spectra of **PcCo** in Fig. 2 showed hypochromic and red changes in the presence of DNA. As concentration of DNA was increased from 0 to 3.5 µM, a strong hyprochromic shift was observed with a small wavelength shift. The considerable hypochromic shifts suggest that a strong interaction occure between **PcCo** and calf thymus-DNA. The absorption spectra of **PcCo** decreased in intensity without changing its conformation after adding different concentrations of calf thymus-DNA. Fig. 2 shows changes in the absorption spectra of **PcCo** during titration with the DNA solution. Dropping in the absorption intensities of **PcCo** can be caused by the existing intercalation binding between the compound and the DNA. This result shows that **PcCo** interacts with calf thymus-DNA through intercalation binding.

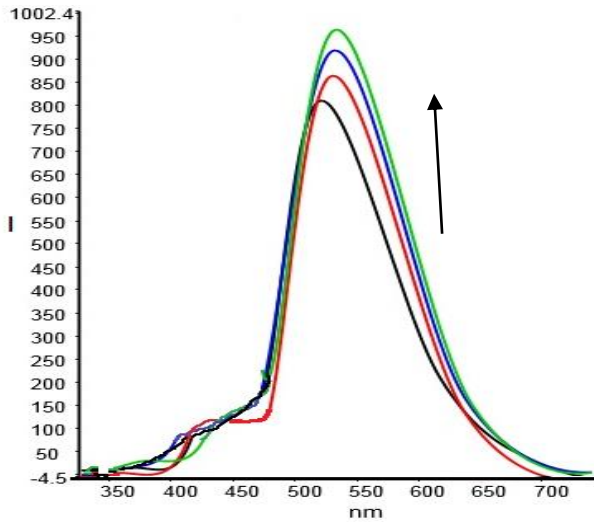

Fig. 3. Fluorescence titration spectra of **PcCo** in the buffer solution. The Arrow indicates intensity change on increasing amount of calf thymus-DNA, where **I** indicates intensity.

Fluorescence titration experiment is also performed to study binding activities between molecular compounds and DNA [30]. Due to fluorescence spectroscopy technique is a prevalent method for DNA-binding study. This technique can give more information for binding between DNA molecule and chemical compounds. As indicated in Fig. 3, calf thymus-DNA + **PcCo** irradiate strong emission in the buffer solution at 7.0 with peak appearance at around 535 nm. On the addition of the DNA sample, a clear increase in emission intensities of **PcCo** was recorded comparing to the original for **PcCo** as shown in Fig. 3. In presence of calf thymus-DNA, **PcCo** gives emission at about 545 nm. The results show that **PcCo** binds strongly to DNA molecule.

3. 2. 2. Cyclic Voltammetry studies

The technique of cyclic voltammetry is largely utilized to investigate binding activities between chemical compounds and DNA. This method also ensures substantial supplementary data to preliminary evaluations of spectroscopic studies [31]. This method is very practical for metal-based molecular compounds because of their reduction and oxidation properties. If a metal based compounds give a reaction with DNA, peak potential and peak current of compound change in the presence of DNA molecule [32].

Cyclic voltammetric technique was carried to comprehend binding activities between calf thymus-DNA and cobalt (II) phthalocyanine (**PcCo**) in the buffer solution and findings are revealed in Fig. 4. In this study, in absence and presence of DNA, cyclic voltammetric experiments were conducted in a buffer. In absence of the DNA, **PcCo** generates a couple of peaks belonging to **PcCo** indicating anodic peak (EPa) and cathodic (EPc). The peak potentials of cathode (EPc) and the peak potentials of anode (EPa) were specified to be 0.65 V, - 0.6 V and − 0.13 V for EPc and 0.9 V, 0.02 V and -0.5 V for EPa for **PcCo** as indicated in Fig. 4a.

On the increasing of DNA with **PcCo**, the cyclic voltammetric peak currents dropped significantly on increasing amount of calf thymus-DNA. The findings indicate that calf thymus-DNA interacts with **PcCo** [33, 34]. The decreasing in voltammetric peak current in the presence of calf thymus-DNA can be based to low diffusion of **PcCo** interaction with calf thymus-DNA molecule. Cathodic peak potential and anodic peak potential for **PcCo** were recorded to be 0.84, -0.36 V for EPc and 0.88 V, 0.15 V, and -0.69 V EPa as indicated in Fig. 4b. All these findings demonstrate that **PcCo** binds to DNA molecule.

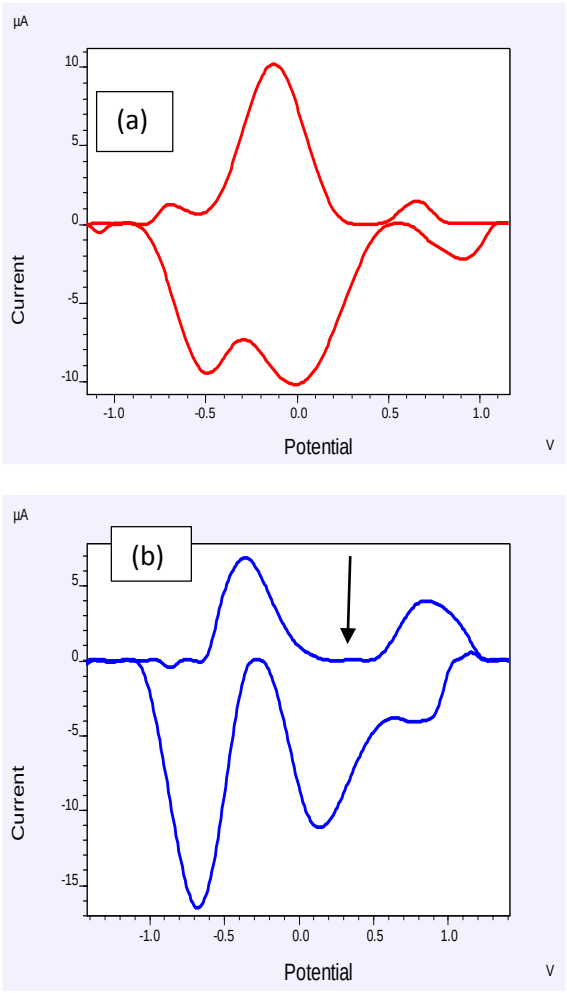

Fig. 4. Cyclic voltammogram of **PcCo** in absence of calf thymus-DNA (a), (red line) and presence of calf thymus-DNA (b) (blue line) of increasing amount of calf thymus-DNA. The arrow indicates decrease in cathodic peak potential on increasing amount of calf thymus-DNA.

3.2.3 Melting point temperature studies

Thermal denaturation study of DNA was conducted to investigate binding activities between **PcCo** and calf thymus-DNA. The melting point temperature of DNA can provide valuable information with regard to the stability of DNA double-helixed with at 260 nm. The intercalation binding of **PcCo** to DNA molecule can enhance to melting temperature of DNA molecule because of strength of binding. In the case of non-intercalative binding of **PcCo** to DNA molecule, the melting point temperature of DNA can drop [35, 36]. In this study, in the absence and presence of **PcCo,** thermal denaturation experiments were performed to specify binding activities between **PcCo** and calf thymus-DNA as indicated in Fig. 5. The melting point temperature for calf thymus-DNA in absence of **PcCo** was found to be 71.67 ºC, and the melting temperature for calf thymus-DNA in presence of **PcCo** was found to be 79.54ºC. These findings indicate that **PcCo** strongly binds to DNA molecule through intercalation binding because of shift in melting point temperature.

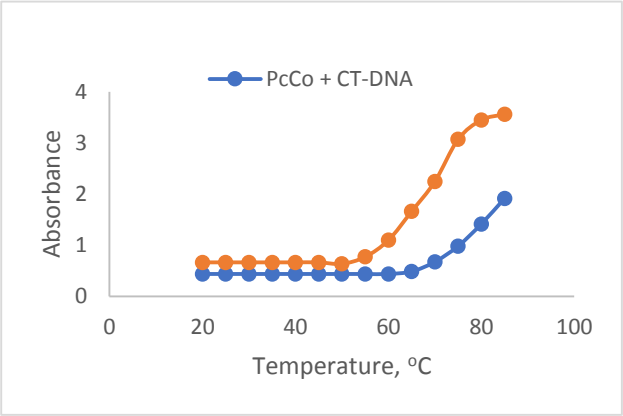

Fig. 5. Melting point temperatures (T_m) of calf thymus-DNA and **PcCo** in a buffer solution.

3.2.4. Viscosity measurements studies

In general, viscosity experiments are conducted to study binding activities between DNA molecule and metal-based compounds. The increase in relative viscosity demonstrates that molecular compounds interact with DNA base pairs via intercalation binding mode, which induces the disintegration and elongation of DNA molecule, and a dropping in relative viscosity shows that compounds bind to DNA base pairs through a non-intercalation binding mode. This disintegration of DNA double-stranded stems from ligands packing between DNA base pairs. The interaction between ligands and DNA base pairs causes a substantial changing in structure of DNA [17, 19]. In this work, viscosity measurements were conducted to invetigate binding activities between **PcCo** and calf thymus-DNA. Fig. 6 indicates there is an increasing in relative viscosity of calf thymus-DNA after successive additions of **PcCo**. These findings demostrate that **PcCo** interacts with DNA through intercalation binding mode.

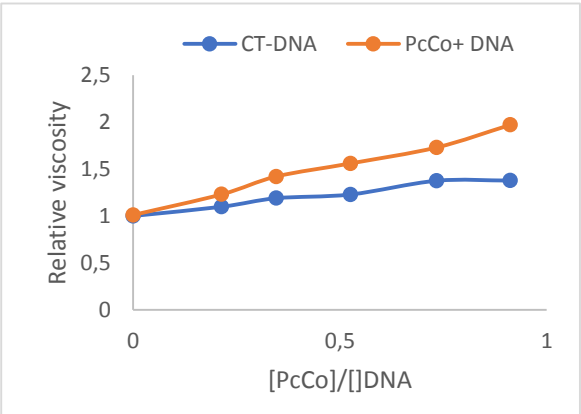

Fig. 6. Realtive viscosity of calf thymus-DNA in the buffer.

3.2.5. Gel Electrophoresis studies

In addition to above studies, gel electrophoresis experiment was conducted to investigate binding activities between **PcCo** and DNA molecule by using calf thymus-DNA. Binding activities between **PcCo** and calf thymus-DNA was studied using agarose gel electrophoresis to determine the impact of varied concentrations of **PcCo** on the DNA. The findings are indicated in Figure 7. These results explicitly show that intensities of DNA bands were dropped after interaction of **PcCo** with DNA when it is compared to the band of calf thymus-DNA control (C). The decreasing in the intensity of DNA bands after interaction of **PcCo** with calf thymus-DNA is considered to be owing to deformation of DNA double-stranded [17, 23, 35, 36, 37].

CT-DNA gel electrophoresis experiment was carried out at room temperature by using synthesized **PcCo**. The interaction of CT-DNA with **PcCo** was monitored by gel electrophoresis using Thermo owl electrophoresis system. The migration of CT-DNA + **PcCo** was monitored after GelRed painting. The electrophoresis experiments clearly revealed that **PcCo** interacts with CT-DNA as there was variation in the bands of lanes 1- 3 compared to the control CT-DNA (C) (**Figure 7**). The interaction efficiency of **PcCo** compared to that of the control CT-DNA (C) originates from their effective CT-DNA binding abilities. As shown in **Figure 7**, the control CT-DNA (lane C) does not display any substantial change of CT-DNA band. **PcCo** Compound (**Figure 7**) (lanes 1, 2, and 3 belong to **PcCo** interact with CT-DNA as compared to control CT-DNA (C). The binding of CT-DNA with **PcCo** results in partial neutralization of DNA bands for 1-3. The presence of a smear in the gel diagram refers to the presence of cleavage. The cleavage potency of the compounds is comparable to that of the control CT-DNA because of their effective DNA interaction ability. The results of gel electrophoresis show that **PcCo** can strongly interact with CT-DNA [38].

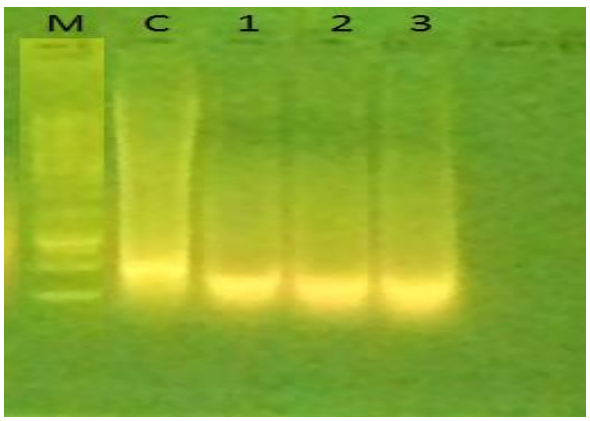

Fig. 7. Agarose gel electrophoresis of CT-DNA in the absence and presence of **PcCo** in a Tris-HCl buffer at pH 7.0. Lane M: DNA Marker; lane C: control CT-DNA; Lanes 1-3: CT-DNA + compound **PcCo** (20 μM), respectively.

4. Conclusion

In this work, the binding activities of **PcCo** with calf thymus-DNA were studied by using UV/Vis absorption spectra and fluoresence emission titration and the results indicated that **PcCo** strongly binds to DNA molecule. Absorption spectra of **PcCo** showed hypochromic and red changes in the presence of DNA. As concentration of DNA was increased from 0 to 3.5 μM, a strong hyprochromic shift was observed with a small wavelength shift. The considerable hypochromic shifts suggest that a strong interaction occure between **PcCo** and calf thymus-DNA. On the addition of the DNA sample, a clear increase in emission intensities of **PcCo** was recorded comparing to the original for **PcCo**. In presence of calf thymus-DNA, **PcCo** gives emission at about 545 nm. The results show that **PcCo** binds strongly to DNA molecule. The large change in melting point temperature of DNA after interaction with **PcCo** also demonstrates intercalation binding. Cyclic voltametric studies show the clear changes in negative peak potentials were recorded on the addition of the DNA also support the intercalation binding of **PcCo** to DNA molecule. The decreasing in voltammetric peak current in the presence of calf thymus-DNA can be based to low diffusion of **PcCo** interaction with calf thymus-DNA molecule. Gel electrophoresis experiments also showed that **PcCo** interacts strongly with the DNA. The cleavage potency of the compounds is comparable to that of the control CT-DNA because of their effective DNA interaction ability. The results were obtained from viscosity measurements also supports intercalation binding between the DNA and **PcCo**. As a result, Co (II) phthalocyanine compound bearing four 4-(2-phenoxy-1, 3-dioxo-2, 3-dihydro-1H-inden-2-yloxy) phthalonitrile group shows strong binding activities with calf thymus-DNA. All the these findings demonstrate that Co (II) phthalocyanine compound can be a promising candidate compound in cancer therapy because of its DNA binding properties.

Acknowledgments

This study was supported by the Commission of Scientific Research Projects of Karabuk University.

References

[1] C.C. Leznoff, A.B.P. Lever, ´´Phthalocyanines Properties and Applications´´ New York, Wiley-VCH, New York, Vol. 4, pp-79-181, 1996.

[2] N.B. McKeown, ´´Phthalocyanines Materials: Synthesis, Structure and Functions´´ Cambridge, MA: Cambridge University Press. 1998.
[3] M.S. Agırtas, A. Altındal, B. Salih, S. Saydam, O. ´´Bekaroglu, Synthesis, characterization, and electrochemical and electrical properties of novel mono and ball-type mettalophthalocyanines with four 9,9-bis(4-hydroxyphenyl)fluorine´´ Dalton Trans., Vol. 40, pp. 3315-3324, 2011.
[4] Y. Peng, H. Zhang, H. Wu, B. Huang, L. Gan, Z. Chen, ´´The synthesis and photophysical properties of zinc (II) phthalocyanine bearing poly (aryl benzyl ether) dendritic substituents´´ Dyes and Pigments, Vol. 87, pp. 10–16, 2010.
[5] M.S. Agırtas, ´´Synthesis and characterization of novel symmetrical pthalocyanines substituted with four benzo [d]>[1,3] dioxol-5-ylmethoxy groups´´ Inorg.chim. Acta., Vol. 360, pp. 2499-2502, 2007.
[6] J-D. Wang, M-J. Lin, S-F. Wu, Y. Lin, ´´15-Bis (2,2,4-trimethylpentoxy)phthalocyanine, a trans-form nonperipheral disubstituted phthalocyanines synthesis by the cross condensation method´´ J. Organomet. Chem., Vol. 691, pp. 5074-5076, 2006.
[7] M. Kandaz, O. Bekaroglu, ´´Synthesis and characterization of novel actakis (2-aminophenoxy and aminophenylsulfanyl)-substituted metallophthalocyanines´´ Chem. Ber./Recueli., Vol. 130, pp. 1833-1836, 1997.
[8] N. Nombona, E. Antunes, T. Nyokong, ´´The sysnthesis and fluorescence behavviour of phthalocyanines unsymmetrically substituted with naphthol and carboxy groupss´´ Dyes and Pigments, Vol.86, pp. 68-73, 2010.
[9] O. Bekaroglu, ´´Ball-type phthalocyanines: synthesis and properties´´ Struct. Bond., Vol. 135, pp. 105-136, 2010.
[10] G. Keser Karaoglan, G. Gumrukcu, A. Koca, A. Gul, ´´The synthesis, characterizatio, electrochemical and spectroelectrochemical properties of a novel, cationic, water-soluble Zn phthalocyanine extended conjugation´´ Dyes and Pigments, Vol. 88, pp. 247-256, 2011.
[11] A.G. Gurek, V. Ahsen, A. Gul, O. Bekaroglu, ´´Synthesis and characterization of a new copper (II) phthalocyaninato substituted with four 15-membered tetraazamacrocycles and its water soluble pentanuclear complexes´´ J. Chem. Soc., Dalton Trans., pp. 3367-3371, 1991.
[12] E. Ben-Hur, R.C. Hoeben, H. Van Ormondt, T.M.A.R. Dubbelman, J. Van Steveninck, ´´Inactivation of retroviruses by phthalocyanines: the effects of sulphonation, metal ligand and fluoride´´ J. Photochem. Photobiol. B: Biol., Vol. 13, pp. 145-152, 1992.
[13] J.D. Spikes, ´´Phthalocyanines as photosensitizers in biological systems Phthalocyanines as photosensitizers in biological systems and for the photodynamic therapy of tumors´´ Photochem. Photobiol., Vol. 43, pp. 691-699, 1986.
[14] J. Ferlay, I. Soerjomatarm, R. Dikshit, R. Eser, C. Mathers, M. Rebelo, D.M. Parkin, D. Forman, F. Bray, ´´Cancer incidence and mortality worlwide: sources, methods and major patterns in Globocan´´ Int. J. Canc., Vol. 136, pp. 359-86, 2015.
[15] R. Gul, A. Badshah, A. Khan, A. Junaid, M.K. Rauf, ´´Preliminary investigation of anticancer activitiy by determining the DNA binding and antioxidant potency of new ferrocene incorporated N,N',N''-trisubstituted phenylgunidines´´ Spectrochim Acta Part A Mol.
Biomol. Spect., Vol. 117, pp. 264-9, 2014.
[16] C. Uslan, B.S. Sesalan, ´´The synthesis, photochemical and biological properties of new silicon phthalocyanines´´ Inorg. Chim. Acta, Vol. 394, pp. 353-62, 2013.
[17] T. Keles, B. Barut, Z. Biyiklioglu, A. Özel, ´´A comparative study on DNA/BSA binding, DNA phorocleavage and antioxidant activities of water soluble peripherally and non-peripherally tetra-3-pyridin-3-ylpropoxy-substituted Mn (III), Cu (II) phthalocyanines´´ Dyes and Pigments, doi: 10.1016/j.dyepig. 2016.12.045, 2017.
[18] S.M. Pradeepa, H.S. Bhojya Naik, B. Vinay Kumar, K. Indira Priyadarsini, A. Barik, S. Jayakumar, ´´Synthesis and characterization of cobalt(II), nickel(II) and copper(II)-based potential photosensitizers: Evaluation of their DNA binding profile, cleavage and Photocytotoxicity´´ Inorg. Chim. Acta, Vol. 428, pp. 138-46, 2015.
[19] B. Zheng, X. Shen, D. Zhao, Y. Cai, M. Ke, J. Huang, ´´Silicon (IV) phthalocyanines substituted axially with different nucleoside moieties. Effects of nucleoside type on the photosensitizing efficiencies and *in vitro* photodynamic activities´´ J. Photochem. Photobiol. B: Biol., Vol. 159, pp. 196-204, 2016.
[20] L. Xodo, S. Cogoi, V. Rapozzi, ´´Photosensitizers binding to nucleic acids as anticanceragents´´ Fut. Med. Chem., Vol. 8, pp. 179-94, 2016.
[21] M.S. Ağırtaş, İ. Gümüş, M. S. İzgi, ´´Designing of new thermo stabile phthalocyanines: synthesis, characterization, and thermal studies´´ Synthesis and Reactivity in Inorganic, Metal-Organic, and Nano-Metal Chemistry, Vol. 42, pp. 1327-1333, 2012.
[22] J. Bolger, A. Gourdon, E. Ishow, J. P. Launay, ´´Mononuclear and Binuclear Tetrapyrido[3,2-*a*:2',3'-*c*:3'',2''-*h*:2''',3'''-*j*]phenazine (tpphz) Ruthenium and Osmium Complexes´´Inorg. Chem., Vol.35, pp. 2937–2944, 1996.
[23] A. S. Azza, M. S. Mohamed, ´´DNA binding, spectrocopic and antimicrobial studies of palladium (II) complexes containin 2,2^{o}-bipyridin and 1-phenypiperazine´´ Spectrochim. Acta Part A Mol. Biomol. Spect., Vol. 96, pp. 586-593, 2012.
[24] M. Özçesmeci, Ö. B. Ecevit, S. Sürgün, E. Humuryudan, ´´Tetracationic fluorinated zinc (ii)

phthalocyanine: Synthesis, characterization and DNA-binding properties´´ Dyes and Pigments, Vol. 96, pp. 52-58, 2013.
[25] P.R. Reddy, A. Shilpa, N. Raju, P. Raghavaiah, ´´Synthesis, structure, DNA binding and cleavage properties of ternaryamino acid Schiff base-phen/bipy C(II) complexes´´ J. Inorg. Bioochem., Vol. 105, pp. 1603-1612, 2011.
[26] A. Apilux, O. Tabata MChailapakul, ´´Electrochemical bihaviors of native and thermal denatured fish DNA in the presence of cytosine derivatives and porphyrin by cyclic voltammetry using boron-drobed diamond electrode´´ Bioelectrochemistry, Vol. 70, pp. 435-439, 2007.
[27] N. Demirezen, D. Tarınc, D. Polat, M. Cesme, A. Golcu, M. Tumer, ´´Synthesis of trimethoprim metal complexes: Spectral, electrochmical, thermal, DNA-binding and surface morp hology studies´´ Spectrochimica Acta Part A, Vol. 94, pp. 243-255, 2012.
[28] X-W. Liu, Y-M. Shen, Z-X. Li, X. Zhong, Y-D. Chen, S-B. Zhang, ´´Study on DNA binding behavior and light switch effect of new coumarin-derived Ru (II) complexes´´ Spectrochim. Acta Part: Mol. Biomol. Specrt., Vol. 149, pp. 150-156, 2015.
[29] C. Ozluer, H. E. Satana Kara, ´´In vitro DNA binding studies of anticancer drug idarubicin using spectroscopic techniques´´ J. Photochem. Photobiol. B., Vol. 138, pp. 36–42, 2014.
[30] A. Özel, B. Barut, Ü. Demirbaş, Z. Biyiklioglu, **"**Invetigation of DNA binding, DNA photocleavage, topoisomerase I inhibition and antioxidant activities of water soluble titanium (IV) phthalocyanine compounds" J. Photoch. Photobiol. B. Biology, Vol. 157, pp.32-38, 2016.
[31] S. Mahadevan, M. Palaniandavar, ´´Spectroscopic and voltametric studies on copper complexes of 2,9-dimethyl-1,10-phenanthrolines bound to calf thymus DNA´´ J. Inorg. Chem., Vol. 37, pp. 693-706, 1998.
[32] M. Sirajuddin, S. Ali, A. Badshah, ´´Drug-DNA interactions and their study by UV-Visible, fluorescence spectroscopies and cyclic voltametry´´ J. Photochem. Photobiol. B. Biology, Vol. 124, pp. 1-19, 2013.
[33] Y.M. Song, P.J. Yang, M.L. Yang, J.W. Kang, S.Q. Qin, B.Q. Lu, ´´Spectroscopic and voltammetric studies of the cobalt (II) complexe of morin bound to cvalf thymus DNA´´ Transition Met. Chem., Vol. 28, pp. 712- 721, 2003.
[34] S.S. Zhang, S. Y. Niu, B. Qu, G.B. Jie, H. Xu, C.F. Ding, ´´Studies on the interaction mechanism between hexakis(imidazole) manganese (II) terephthalate and DNA and preparation of DNA electrochemical sensor´´ J. Inorg. Biochem., Vol. 99, pp. 2340-2347, 2005.
[35] M.T. Cater, M. Rodriguez, A.J. Bard, ´´Voltammetric studies of the interaction of metal chelated with DNA. 2. Tris-chelated complexes of cobalt (III) and iron (II) with 1,10-phenanthroline and 2, 20- bipryridine´´ J. Am. Chem. Soc., Vol. 111, pp. 8901-8911, 1989.
[36] P. Pravin, P.P. Utthra, G. Kumaravel, N. Raman, ´´Effective DNA binding and cleavage tendencies of malonic acid coupled transition metal complexes´´ J. Mol. Struc., Vol. 1123, pp. 162-70, 2016.
[37] L. Hassani, F. Hakimian, E. Safaei, Z. Fazzeli, ´´Antibacterial effect of cationic porphyrazines and anionic phthalocyanine and their interaction with plasmid DNA´´ J. Mol. Struc., 1052, 221-227, 2013.
[38] M.M. Al-Mogren, M.A. Alaghaz, E.A. Ebrahem**,** ´´Synthesis, spectroscopic, molecular orbital calculation, cytotoxic, molecular docking of DNA binding and DNA cleavage studies of of transition metal complexes with N-benzylidene–N´-salicylidene-1,1-diaminopropane´´ Spectrochim. Acta Part A Mol. Biomol. Spectr., Vol. 114, pp. 695-707, 2013.