

Synthesis of Tetra(*m*-nitrophenyl)porphyrin as Sensing Material for Recognition of Lead(II) Ion

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The monitoring of lead in the environment has gained importance because lead(II) ion is a toxic metal ion causing adverse environmental and health problems. In this paper, we reported the synthesis of 5,10,15,20-tetra(*m*-nitrophenyl)porphyrin as a sensing material aimed at the recognition of lead(II) ion. The synthesis and ion binding characteristics of the 5,10,15,20-tetra(*m*-nitrophenyl)porphyrin were investigated using FTIR, ¹H NMR, and UV-Vis spectroscopy. The binding of lead(II) ion to the 5,10,15, 20-tetra(*m*-nitrophenyl)porphyrin was found to give a new peak in the absorption spectra at 473.35 nm along with two isobestic points at 483 nm and 465 nm, suggesting porphyrin complex formation of 5,10,15, 20-tetra(*m*-nitrophenyl)porphyrin-lead(II). The calculated limit of detection for lead(II) ion using 5,10,15, 20-tetra(*m*-nitrophenyl)porphyrin is 3.67×10^{-4} M and the association constant, K_{ass} is $1.56 \times 10^2 \text{ M}^{-1}$. Herein, it was proposed that a new coordination compound was formed between lead(II) ion with 5,10,15, 20-tetra(*m*-nitrophenyl)porphyrin through nitro group chromophore.

Key words: Lead(II) ion; porphyrin; sensing; chromophore

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Until the 1970s, lead was commonly added to gasoline, house paint and cosmetic as an additive for high-performance engines, pigments and whitens the skin, respectively. However, after the widespread application of lead compound in industries, many health issues especially, childhood lead poisoning was started [1]. People can be exposed to lead through breathing, drinking, eating and contact with dust containing the lead compound. Lead mimics biologically helpful minerals such as calcium, iron, and zinc. Due to these properties, lead preferentially replaces all these metals in biochemical reactions in the human body. When lead displaces other metals in the protein molecules, a change in the protein shape occurs that interferes with the function of the protein. This health issue needs to be overcome

with the further development of lead monitoring devices or sensors.

To become a chemical sensor for monitoring in the field, the chemical sensor should not be expensive, providing notable sensitivity, rapid response time and simple in fabrication and handling [2]. In terms of the sensor material, it needs to have high sensitivity, low detection limits, easy to synthesize, suitable for a variety of analytes and stable towards heating, pH, and light [3]. Owing to all these criteria, various material structures have been used in chemical sensors, depending on the nature of the quantity to be measured, ranging from single crystals to amorphous polymers [4]. Other than that, for analytical chemistry applications using spectroscopy, chromatography, and

electroanalysis, macrocycles and supramolecules have gained much attention due to the ability of these materials to convert the non-electric information into electric signals [5].

Additionally, supramolecule of porphyrin is naturally having strong and deep colour with high wavelength, ease the binding analysis using spectroscopic methods. In the chemical sensor field, there are only a few papers that emphasize on the application of meso(nitrophenyl)porphyrin. In 2012, Kangwanwong and co-workers synthesized this type of porphyrin as fluorescence chemosensor for ruthenium(III) ion with a detection limit of 0.89 μM [6]. Besides that, the meso-substituted porphyrin derivative was also synthesized and incorporated into nafion film for the detection of ammonia gas and subsequently, optochemical sensing using the UV-Vis spectrometer [7]. Moreover, a thin film of it was also employed for electrochemical sensing of hydrochloric acid gas using electrochemical workstation [8]. As well in this study, 5,10,15,20-tetra(*m*-nitrophenyl)porphyrin (afterward written as tetra(*m*-nitrophenyl)porphyrin) was synthesized and applied for the first time for lead(II) ion sensing.

EXPERIMENTAL

All solvents, chemicals, and reagents were purchased from Merck and Sigma-Aldrich

international chemical companies and used without further purification. IR and UV-Vis spectra were recorded on a PerkinElmer Frontier FTIR spectrometer using KBr pellet technique and a PerkinElmer Lambda 25 UV-Vis spectrometer, respectively. NMR spectra used in the characterization of products were recorded on a Bruker DPX-400 MHz NMR spectrometer using deuterated chloroform as a solvent. MALDI-TOF spectrometer from model AB Sciex TOF/TOF 5800 was used for mass analysis using α -cyano-4-hydroxycinnamic acid as a matrix.

In the general procedures for preparation of tetra(*m*-nitrophenyl)porphyrin (Figure 1), 3-nitrobenzaldehyde (0.04 mol), pyrrole (0.04 mol) and propionic acid (150 ml) were placed in a round bottom flask. Then, the resulting mixture was stirred and heated up to 180°C at reflux for 1 h. After completion of the reaction, the crude product was filtered and washed with methanol (3×20 ml). Further purification was done by silica column using chloroform as a solvent.

General Spectroscopic Method

A stock solution of lead(II) ion was prepared from lead(II) nitrate along with tetra(*m*-nitrophenyl)porphyrin with concentration 0.01 M and 1×10^{-4} M, respectively. Both of the stock solutions were freshly prepared using dimethylsulphoxide (DMSO) as a solvent. After that, 0.1 ml of lead(II) ion solution was titrated into 10 ml of tetra(*m*-

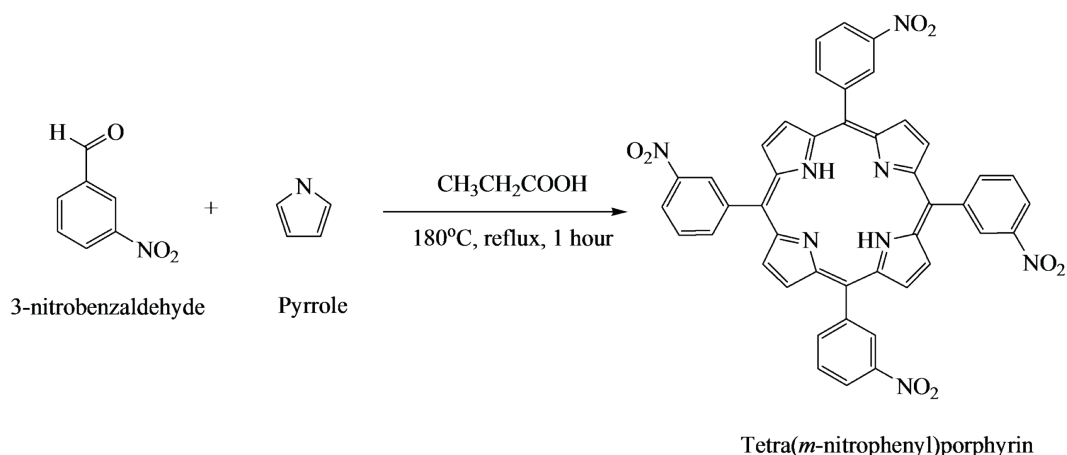


Figure 1. Synthesis of tetra(*m*-nitrophenyl)porphyrin.

nitrophenyl)porphyrin solution and the changing in the absorption was recorded using UV-Vis spectrometer. The titration step was repeated for 30 times, and the collected data was used to determine the limit of detection and association constant (K_{ass}). The detection limit of lead(II) ion that can be recognized by tetra(*m*-nitrophenyl)porphyrin can be determined using Equation 1 [9]:

$$\text{Limit of detection} = 3.3 \times \frac{\text{Standard error regression slope}}{\text{coefficient}} \quad (\text{Equation 1})$$

Additionally, the standard error of regression and slope coefficient were calculated using the LINEST function in *Excel* to find the best-fit line, with concentration of lead(II) ion and tetra(*m*-nitrophenyl)porphyrin absorption intensity as the *x* and *y* variables. Meanwhile, binding or association constant, K_{ass} of tetra(*m*-nitrophenyl)porphyrin-lead(II) complex is calculated using the Benesi-Hildebrand method (Equation 2) [10]:

$$\frac{\Delta A}{b} = \frac{\Delta \varepsilon K_{\text{ass}} [G][H]}{1 + K_{\text{ass}} [G]} \quad (\text{Equation 2})$$

Referring to the above equation, ΔA is the change in absorbance, K_{ass} is the association constant, $\Delta \varepsilon$ is molar absorptivity, $[G]$ is the concentration of guest, and $[H]$ is a concentration of host. In this study, the guest was lead(II) ion and the host was tetra(*m*-nitrophenyl)porphyrin. Subsequently, both K_{ass} and $\Delta \varepsilon$ are constants. Therefore the SOLVER function in *Excel* was used.

RESULTS AND DISCUSSION

The synthesis of tetra(*m*-nitrophenyl)porphyrin is summarized in Figure 1. It began with the electrophilic substitution reaction with condensation of 3-nitrobenzaldehyde and pyrrole. Repeated condensations formed a rapidly equilibrating mixture of short pyrrole-aldehyde chains. As a catalyst and solvent, propionic acid was used instead of a strong acid in order to prevent the dimerization of pyrrole with the

increasing of electrophilicity of benzaldehyde. Hence, it becomes more likely for the pyrrole to attack it instead of undergoing dimerization [11]. In the FTIR spectrum (Figure 2), the absence of aldehyde stretching peaked at frequency $\sim 1725 \text{ cm}^{-1}$, indicated the complete reaction between 3-nitrobenzaldehyde and pyrrole. Other than that, the sharp and small peak presented at 3325 cm^{-1} showed a secondary amine from the porphyrin pyrrolic ring. Moreover, the two peaks at 1525 cm^{-1} and 1344 cm^{-1} signify the aromatic nitro compound (conjugated) for the asymmetric and symmetric stretches. The appearance of a hydroxy peak at 3396 cm^{-1} was attributed to the water in the air.

For further characterization of tetra(*m*-nitrophenyl)porphyrin, the ^1H NMR spectroscopy was used (Figure 3). In the porphyrin structure, the amine group, $-\text{NH}$ in the porphyrin ring was very important to indicate the complete reaction between 3-nitrobenzaldehyde and pyrrole. In the further characterization of the tetra(*m*-nitrophenyl)porphyrin using ^1H NMR spectrometer, a low chemical shift at -2.809 ppm with singlet band showed the presence of H_a proton. Owing to the anisotropic effect from the ring current in the system, this proton was shielded, resulting in the lowering of chemical shift. For the same protons at beta position of the porphyrin pyrrolic ring, H_b and H_g , a doublet band at high chemical shifts of 8.599 ppm and 8.587 ppm were present, which showed the presence of a neighbouring proton (H_b proton being a neighbour for H_g and H_g proton being a neighbour for H_b). On account of the anisotropic effect of the conjugated porphyrin system, H_b and H_g protons were deshielded, thus observed at a higher chemical shift value compared to H_a . H_c and H_e bands were present at 8.760 ppm and 8.739 ppm , respectively. Other than that, these two protons being neighbours around H_d protons, resulted in the triplet bands at 8.047 ppm , 8.029 ppm , and 8.009 ppm . The ortho protons, H_f , had no neighbouring proton, thus showed a singlet band at 8.847 ppm . These complete NMR analyses thus confirmed that tetra(*m*-nitrophenyl)porphyrin was indeed synthesized.

In addition to the FTIR and ^1H NMR analysis, the mass analysis was carried out using MALDI-

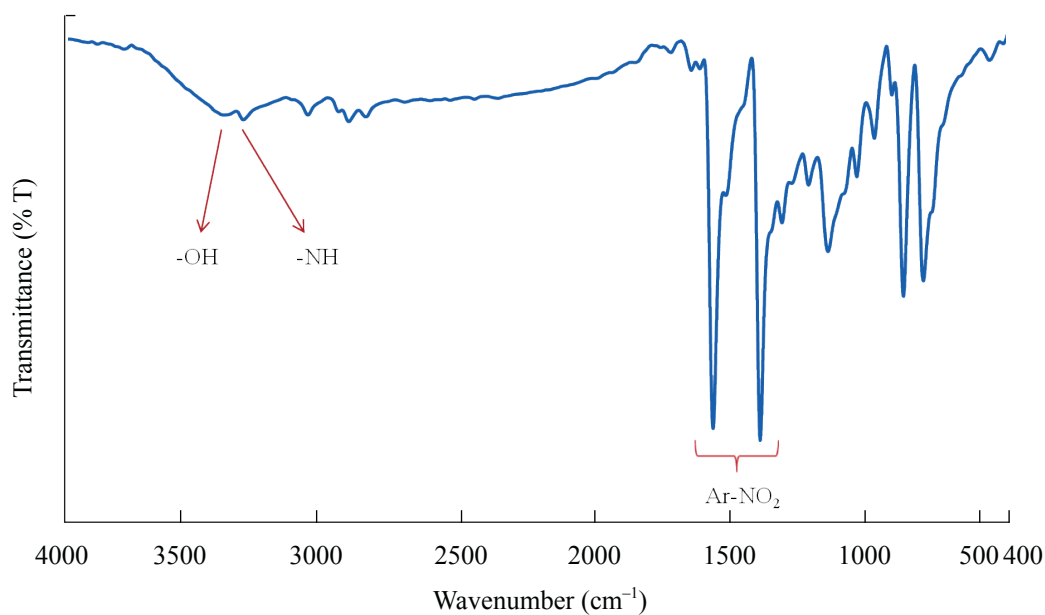


Figure 2. FTIR spectrum of tetra(*m*-nitrophenyl)porphyrin.

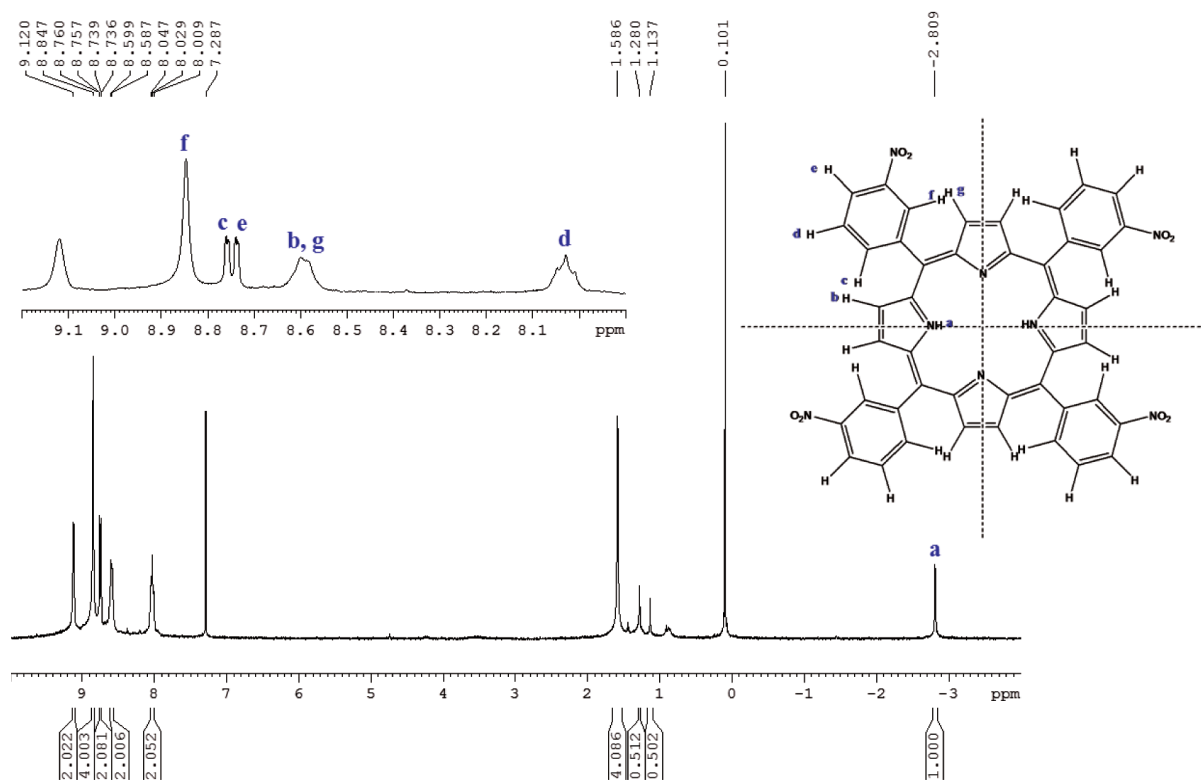


Figure 3. ¹H NMR spectrum of tetra(*m*-nitrophenyl)porphyrin.

TOF spectrometry. The MALDI-TOF mass spectrum of synthesized tetra(*m*-nitrophenyl)porphyrin is shown in Figure 4. The actual mass of tetra(*m*-nitrophenyl)porphyrin was 794.19 m/z. Using α -cyano-4-hydroxycinnamic acid as a matrix, the synthesized tetra(*m*-nitrophenyl)porphyrin was analyzed and found in the form of protonated $(M+H)^+$ with mass 795.1725 m/z. The high peak at this mass indicated the purity of the synthesized product.

To study the binding of lead(II) ion with tetra(*m*-nitrophenyl)porphyrin, (UV-Vis spectrometer was used with DMSO as a solvent to study the binding of lead(II) ion with tetra(*m*-nitrophenyl)porphyrin. DMSO as a polar aprotic solvent did not have unshared pair of electrons. Therefore it decreased the competition between solvents and lead(II) ion molecule to interact and bind with the tetra(*m*-nitrophenyl)porphyrin, as compared to using polar

protic solvents such as water and methanol that had unshared electron pairs. According to UV-Vis spectra in Figure 5, tetra(*m*-nitrophenyl)porphyrin have five fingerprint peaks with the Soret band at 429.34 nm and four Q bands at 515.24 nm, 548.70 nm, 588.78 nm and 644.84 nm. After titration of lead(II) ion, the intensity of these peaks was decreased without shifting or diminishing. In addition, a new peak appeared at 473.35 nm with two isobestic points at 483 nm and 465 nm which indicated the formation of the new complex of tetra(*m*-nitrophenyl)porphyrin-lead(II).

Figure 6 presents the proposed coordination binding of the new complex. Based on the study by Magyar and co-workers, in almost all cases, lead(II) ion commonly has 5, 6 or 8 coordination number [12]. Therefore, in this study, it was suggested that when the concentration of lead(II) ion was low compared to tetra(*m*-nitrophenyl)

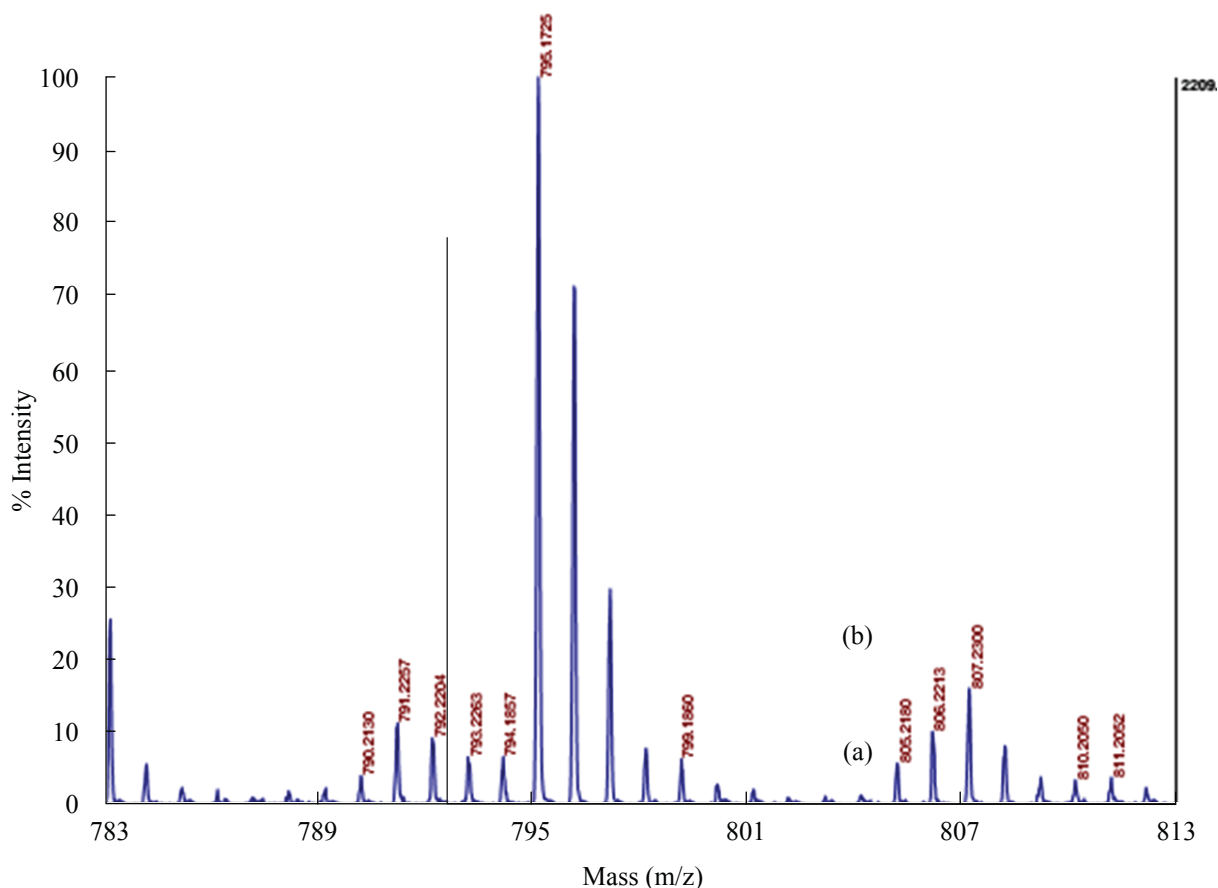


Figure 4. MALDI-TOF mass spectrum of tetra(*m*-nitrophenyl)porphyrin.

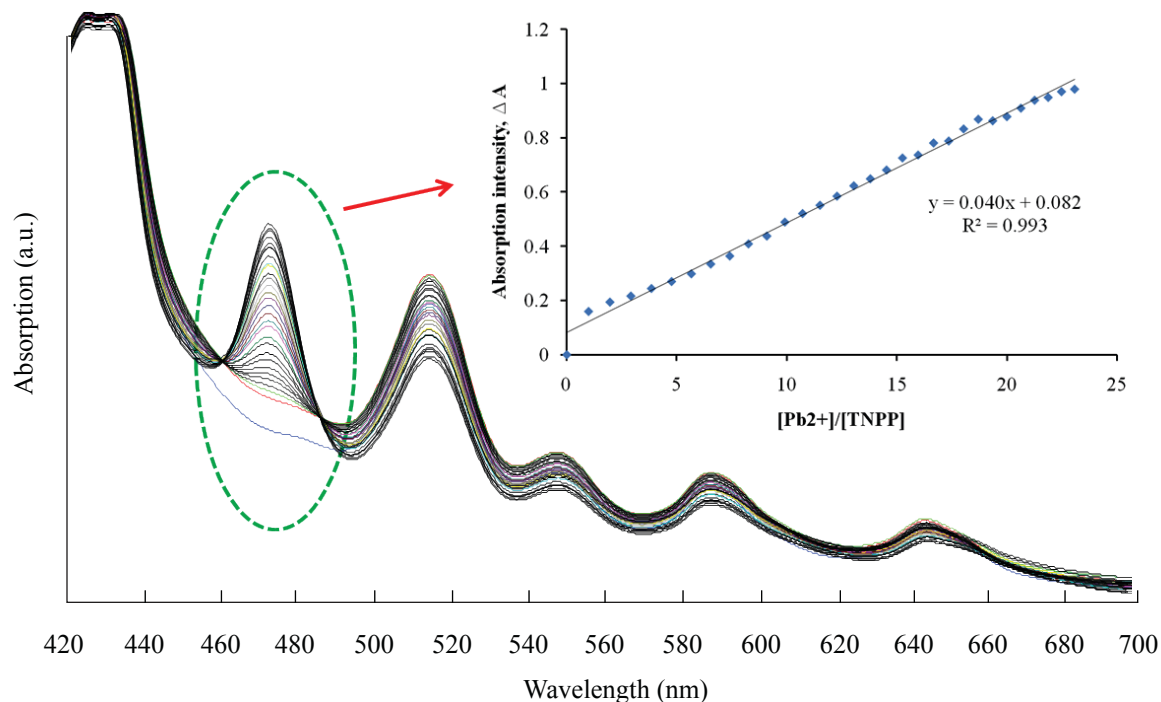


Figure 5. UV-Vis spectra for titration of tetra(*m*-nitrophenyl)porphyrin with lead(II) ion.

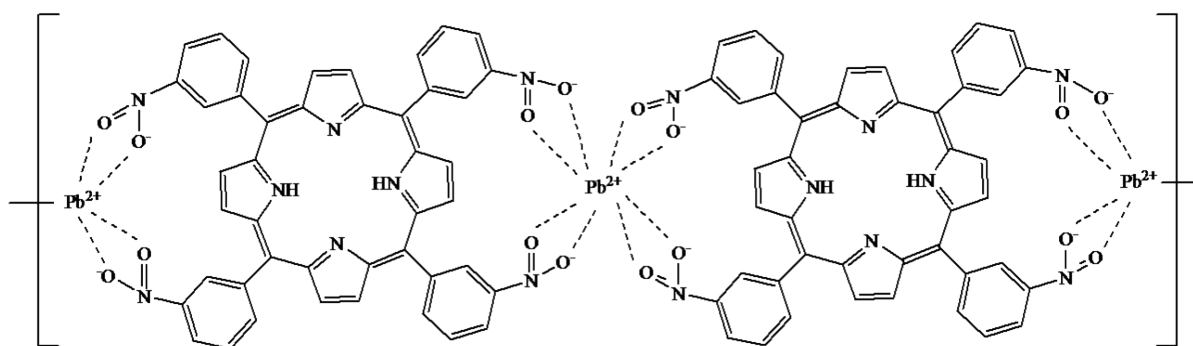


Figure 6. Proposed coordination binding between tetra(*m*-nitrophenyl)porphyrin with lead(II) ion.

porphyrin concentration, the coordination number was 8. However, when the concentration of the lead(II) ion was increasing, the coordination number became 6 or 5. The chromophore of the nitro group at the side of the tetra(*m*-nitrophenyl) porphyrin was bonded to the lead(II) ion causing the existence of the new peak. Lead(II) ion had ionic radius ~ 0.133 nm, and it was considered as big, compared to the porphyrin ring. Even though, the amine group on the inside of porphyrin

ring was preferable to form the coordination bonding with lead(II) ion than the nitro group at the aromatic ring; the limited space or size of the porphyrin ring might prevent the formation of metalloporphyrin. This occurrence could be evidenced by the presence of four Q band peaks even though after binding with lead(II) ion. The normal formation of metalloporphyrin was noticed by the disappearance of two peaks at Q band region (500–750 nm).

The association constant or so-called binding constant is a special case of equilibrium constant and is the inverse of the dissociation constant. In other words, the association constant, K_{ass} is a numerical constant used to describe the bonding affinity and molecular binding strength of two molecules at equilibrium. Therefore in this research described the binding strength between tetra(*m*-nitrophenyl)porphyrin and lead(II) ion. Using the UV-Vis titration data, the association constant for coordination bonding between tetra(*m*-nitrophenyl)porphyrin with lead(II) ion was calculated as K_{ass} , $1.56 \times 10^2 \text{ M}^{-1}$ and the limit of detection is $3.67 \times 10^{-4} \text{ M}$. With these data, tetra(*m*-nitrophenyl)porphyrin showed a remarkable potency to be used as the lead(II) ion sensing materials in many fields such as agriculture, cosmetic, food, coating, and others.

CONCLUSION

In summary, this new application of tetra(*m*-nitrophenyl)porphyrin as the lead(II) ion sensing materials opened the door for further developments of porphyrin molecule for the monitoring of poisonous heavy metals especially lead element. The main feature of this tetra(*m*-nitrophenyl)porphyrin was its ability to recognize and bind with lead(II) ion and transform the binding events into the electric signals that could be detected using the UV-Vis spectrometer. This attractive property, coupled with its ease of synthesis and quick response, led us to suggest that it might prove beneficial in preventing lead(II) ion poisoning in humans.

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