XXI Brazilian Meeting on Inorganic Chemistry
X Brazilian Meeting on Rare Earths
III Workshop on Theoretical Bioinorganic Chemistry
Belo Horizonte, September 12 - 15th 2024

Study of the binding mechanism of a cationic porphyrin and its zinc complex with human serum albumin via fluorescence quenching

Hanya Cristina Moura Almeida, Pedro Fonseca-Pinheiro, Nathany Isabelly Dias Vieira, Dayse Carvalho da Silva Martins*

Departamento de Química, ICEx/UFMG

*E-mail: hanya3105adm@gmail.com; *daysequimica@ufmg.br*

Thematic Area: Biological Inorganic Chemistry.

Keywords: biomolecules, metalloporphyrins, photodynamic therapy.

The use of porphyrins as photosensitizers in photodynamic therapy (PDT), as an alternative to the methods currently used to treat cancer, has been growing in the research field and showing positive results. Research has shown that photosensitizers are transported through the cardiovascular system mainly by human serum albumin (HSA), which is the most abundant protein in human blood plasma.[1] In this work, the binding between HSA and the free base porphyrin 5,10,15,20-tetrakis(N-ethyl-pyridinium-3-yl) porphyrin (H_2P), as well as its complex with Zn^{2+} (ZnP) at three different temperatures, was studied using fluorescence quenching. There are two different mechanisms of fluorescence quenching. The static mechanism occurs when the quencher forms a non-fluorescent complex with the fluorophore before excitation, and the dynamic mechanism occurs when the quencher interacts with the fluorophore after excitation. For this study, after each addition of porphyrin to the HSA solution, the samples were excited at 280 nm and scanned spectrophotometrically from 290-500 nm. Using the obtained data, it was possible to calculate the suppression constant (K_{sv}), the fraction of initial fluorescence accessible to the suppressor (f_a), the suppression constant for the fraction accessible to the suppressor (K_a), the number of binding sites (n), and the association constant (K_b)[2] (Table 1). The increase in temperature for the HSA-[H_2P] bond resulted in a decrease in the average K_{sv} values indicating a static mechanism. Regarding the HSA-[ZnP] bond, the increase in temperature increased the average K_{sv} values, associated with a dynamic mechanism. There was an increase in K_b as the temperature increased, indicating an increase in the stability of the derivative formed between HSA and both photosensitizers. The number of binding sites calculated is approximately 1, showing that there is only one porphyrin molecule bound to the albumin in both reactions. It is possible to verify the potential of the binding between HSA and cationic porphyrin compounds since all K_{sv} values are in the order of 10^4, exactly the ideal range of K_{sv} for the porphyrin to be transported, which means the protein can transport the studied cationic porphyrins through the cardiovascular system.

Table 1 - Values of K_{sv}, f_a, K_a, K_b, and n associated with the bound of porphyrins with HSA.

	K_{sv} 25°C ($\times 10^4$ mol^{-1}.L)	K_{sv} 30°C ($\times 10^4$ mol^{-1}.L)	K_{sv} 35°C ($\times 10^4$ mol^{-1}.L)	f_a	K_a 25°C ($\times 10^4$ mol^{-1}.L)	K_a 30°C ($\times 10^4$ mol^{-1}.L)	K_a 35°C ($\times 10^4$ mol^{-1}.L)	K_b 25°C ($\times 10^4$ mol^{-1}.L)	K_b 30°C ($\times 10^4$ mol^{-1}.L)	K_b 35°C ($\times 10^4$ mol^{-1}.L)	n
H2P	9.11	8.5	8.43	1.62 at 25C 1.11 at 30C 0.41 at 35C	3.79	5.46	10.33	28.8	28.93	97.87	1
ZnP	7.09	7.25	7.55	N/A	N/A	N/A	N/A	19.68	23.10	24.22	1

Acknowledgments: CAPES, CNPq, FAPEMIG, FINEP, INCT-CiMOL, PRPq/UFMG.

References

[1] C. André, Y.C. Guillaume, Journal of Inorganic Biochemistry, **63**, 503-508 (2004).

[2] B.M. Mariete, S.F. Douglas, C.F. Kalil, B. João Raul, S.N. Helena, R. Vagner, Química Nova, **38**, 227-232 (2015).