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Mössbauer Spectroscopic Studies on Concentrated Protoporphyrin IX Iron(II) Solutions

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Abstract

^{57}Fe Mössbauer parameters are presented for protoporphyrin IX iron(II) frozen solutions These are interpreted as evidence for changes in the electronic environments around the iron(II) centres when the metalloporphyrin molecules aggregate in solution.

Introduction

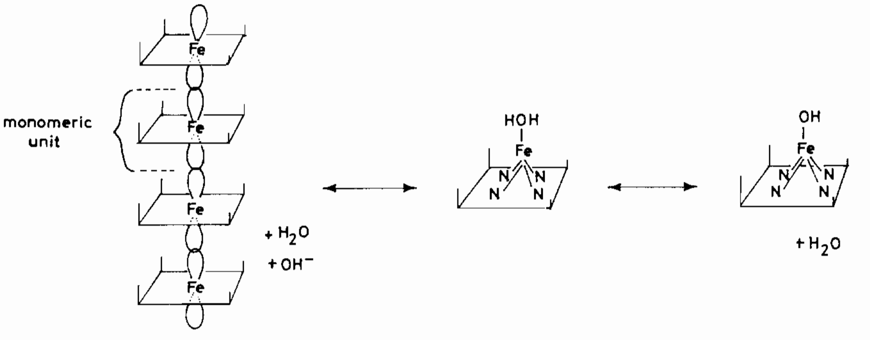
Over the last few years we have undertaken studies of water soluble ironII/III porphyrins over the pH range 3–14+ using mainly electronic absorption spectroscopy at room temperature and Mössbauer spectroscopy on frozen solutions at 80 K [1–8]. To enable dilute solutions to be studied iron porphyrins enriched with ^{57}Fe were used for the Mössbauer spectroscopy experiments One of the reasons for choosing to use ^{57}Fe in the studies was to avoid problems associated with aggregation of the porphyrins. The phenomenon of aggregation in porphyrins and metalloporphyrins has been well documented and a useful review has appeared [9].

We have previously reported studies on frozen solutions of protoporphyrin IX iron(III), hereinafter written as [PPIXFe(II)], [2, 4]. In these studies we used PPIXFe(II) prepared from a 90% ^{57}Fe enriched $FeCl_2$ solution. We found evidence for three different iron(II) sites (see Fig. 1) in the pH range 7 to 14+ [2, 4]. These were species **A**, an intermediate spin iron(II) site that we ascribed to a polymer species; species B, a high spin iron(II) site that we have ascribed to a monomeric species, characterised by Mössbauer parameters $\delta = 0.93(7)$ mm s^{-1} and $\Delta = 2.25(4)$ mm s^{-1}, and species C, a second high spin iron(II) site where the iron atom was further out of the plane (with $\delta = 1.01(2)$ and $\Delta = 4.11(3)$ mm s^{-1}).

Recently we have had reason to examine naturally occurring PPIXFe(II) extracted from bovine blood in concentrated frozen solutions The results observed differ from those of our previous studies and are reported here.

Results and Discussion

The results of our Mössbauer spectroscopic studies are reported in Table 1 and in Fig 2 The



Species A
Exists in solid, and is dominant in solutions over the pH range 7-14

Species B
Exists from pH 10·3 to above pH 14 (but only in small amounts)

Species C
Exists from pH 12 to 14 and is dominant above pH 14

Fig 1. Species postulated to be present in solutions of PPIXFe(II) at high pH.

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TABLE I. Mössbauer Parameters of Frozen Solutions of Protoporphyrin IX Iron(II) Solutions

pH	mg of PPIXFe(II) per 3 ml	γ (mm s^{-1})^a	Δ (mm s^{-1})	Γ (mm s^{-1})	%Absorption area	Reference
11.95	5	0.52(2)	1.41(2)	0.20(2)	67(3)	2
		0.93(7)	2.27(13)	0.27(7)	11(3)	
		0.98(2)	4.15(3)	0.20(2)	22(2)	
12.0	100	0.59(2)	1.47(2)	0.24(2)	100(3)	This work
12.4	150	0.55(1)	1.57(1)	0.25(2)	100(4)	This work
12.5	200	0.60(1)	1.63(1)	0.18(1)	100(3)	This work
12.5	200	0.60(1)	1.64(2)	0.19(1)	100(2)	This work
12.7	200	0.60(2)	1.65(2)	0.19(1)	100(3)	This work
13.0^b	200	0.61(1)	1.58(1)	0.20(1)	100(4)	This work
13.0^{c,d}	200	0.63(1)	1.61(2)	0.24(2)	47(4)	This work
		1.04(1)	4.49(1)	0.17(1)	53(3)	
>14	5	0.52(2)	1.40(4)	0.24(1)	43(2)	2
		1.01(3)	2.25(4)	0.18(3)	15(2)	
		1.01(1)	3.97(1)	0.18(1)	43(1)	

^{a}Half width at half height. ^{b}The spectrum at this pH showed a trace of a second Fe(II) site with a Δ of around 4.4(2) mm s^{-1}. ^{c}The spectrum at this pH showed a trace of a third Fe(II) site with a Δ of around 2.4(1) mm s^{-1}. ^{d}Greater than pH 13.0.

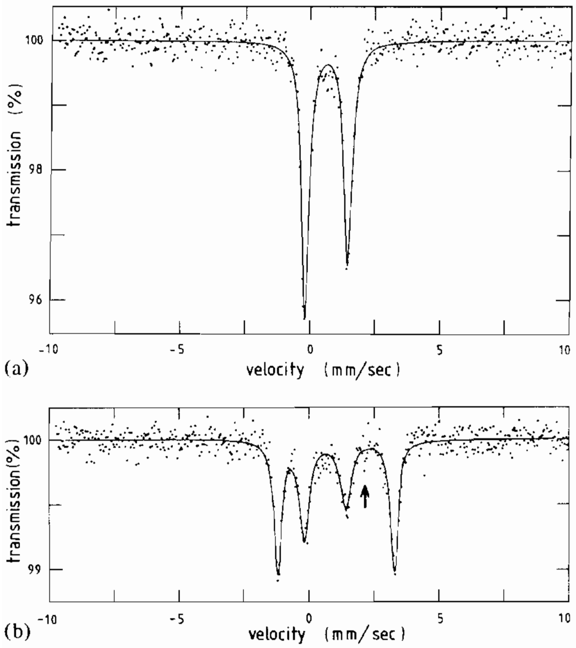

Fig. 2. Mössbauer spectra of protopoprhyrin IX iron(II) (containing natural abundance ^{57}Fe). (a) frozen solution pH 12.5; (b) frozen solution pH 13+. Arrow indicates some evidence for a site with Δ around 2.4(1).

site (Fig. 1) which we previously ascribed to, an $S = 1$ intermediate spin iron(II) with a $\Delta = 1.41(2)$ mm s^{-1} and $\delta = 0.52(2)$ mm s^{-1} is seen to show different parameters in more concentrated solutions. The quadrupole splitting is seen to increase with concentration of PPIXFe(II), the isomer shift also appears to increase.

The solutions which gave rise to these spectra are very concentrated indeed, but were solutions before freezing, and contained no undissolved solid. The obvious explanation for the change in the Mössbauer parameters with concentration of PPIXFe(II) is that aggregation has taken place, and that this phenomena is manifest as a change in the electronic environment at the iron centre. Thus we suggest that aggregation changes the electron density on the porphyrin ring by porphyrin–porphyrin interaction (possibly due to electronic interaction between the porphyrin rings via some kind of orbital overlap). Such interaction will withdraw electron density from the porphyrin ring and is apparent as an increase in Δ. It should be noted that in the structure of TPPFe(II) [10] the porphyrin–porphyrin distances are around 3.5 Å and the ^{57}Fe Mössbauer parameters are similar to those found for site A (Fig. 1) in solutions containing 100 mg PPIXFe(II) (We previously erronously gave the Fe•••Fe distance in solid TPPFe(II) [2] as 3.8 Å, due to the statement given in ref. 11, which contains compounds isostructural to TPPFe(II). Clearly the Fe–Fe distance is much greater than this. The authors obviously must have meant the porphyrin–porphyrin distance.) The fact that TPPFe(II) has Mössbauer parameters similar to those found in dilute PPIXFe(II) solutions suggests that either little porphyrin–porphyrin electronic interactions take place in the solid state or some interactions take place even in dilute solution. In TPPFe(II) ref. 10, $\Delta = 1.51(1)$ mm s^{-1} which is larger than what we observe for dilute PPIXFe(II) solutions [1], this may be evidence of porphyrin–porphyrin interactions. There is a line up of pyrrole C atoms in adjacent rings in the packing diagram in the isostructural CuTPP [11], though the

C···C distance between the porphyrins is long at 3.5 Å. We have previously suggested [2] that species A is a polymer species possibly caused by Fe···Fe interactions through the d_{z^2} orbitals. In the more concentrated solutions studied here, we expect that such polymers would aggregate and that porphyrin–porphyrin interactions between the polymers would take place. By necessity from the assumptions that either no porphyrin–porphyrin interactions take place in TPPFe(II) or that they happen even in dilute solutions, then for such interaction the porphyrin–porphyrin distances must be equal to or less than 3.5 Å. So in solution either the porphyrin–porphyrin interactions must be of the order of 3.5 Å, or the overlap between the PPIX rings could be via the vinyl groups (and would still have to be around 3.5 Å apart).

Other changes are apparent in the Mössbauer data. Previously we found a species defined as B (Fig. 1) with a $\Delta = 2.25(2)$. In these more concentrated solutions we only find slight evidence for this species with a $\Delta \sim 2.4(1)$. The fact that such a relatively small amount of this site is apparent in concentrated solutions we take as evidence for it being a monomeric five coordinate Fe(II) site with one axial H_2O ligand, which is not favoured by aggregation.

We find that a species with similar though larger Mössbauer parameters to those we reported for species C (Fig. 1) exists above pH 13.00. These larger parameters may also be rationalised by aggregation using similar arguments to those presented for the intermediate spin iron(II) site (*i.e.* porphyrin–porphyrin electronic interactions). The Δ of 4.49(1) mm s^{-1} is the largest yet recorded for a high spin iron(II) porphyrin species. There is a greater proportion of this site present than in the dilute solutions, this could be due to enhanced formation from species B on aggregation.

Conclusions

Evidence for aggregation in metalloporphyrins has previously been reported using Electronic absorption spectroscopy, and NMR studies [9]. Thus it is perhaps not surprising that changes in the Mössbauer parameters should be associated with aggregation of metalloporphyrins.

Such aggregates with their modified iron(II) electronic environments might be expected to interact with ligands differently to monomeric iron(II) porphyrin species.

Experimental

Haematin (from Bovine blood) was purchased (from Sigma) and used without further purification. The frozen solutions used for the Mössbauer spectroscopic experiments were made up by dissolving the required number of mgs haematin in 0.5 ml 1 N NaOH solution and then diluted to 3 ml with distilled water. The pH was adjusted using the appropriate calculated amount of 2 N NaOH for pH's > 12. The solutions under an N_2 atmosphere were reduced using a few drops of a concentrated sodium dithionite solution.

The concentrations studied in this work were dictated by solubility of PPIXFe(II), and also by having enough naturally occurring complex present to enable ^{57}Fe Mössbauer spectra to be collected in 1–3 day runs (200000 to 10^6 counts per channel), with reasonably narrow line widths.

The Mössbauer spectra were recorded on an instrument previously described [12]. Solutions were encapsulated in nylon cells under N_2 atmosphere and quench frozen in liquid N_2, then transferred to the cryostat (78 K). The source was ^{57}Co (25 mCi) in rhodium (Radio chemical centre Amersham), at 20 °C. The spectrometer was operated in a saw tooth mode and the spectra computer fitted. The spectrometer was calibrated with 25 μM thick natural iron reference absorber. All isomer shifts are referred to this as zero shift.

References

1 J. Silver and B. Lukas, *Inorg. Chim. Acta, 78,* 219 (1983).
2 J. Silver and B. Lukas, *Inorg. Chim. Acta, 80,* 107 (1983).
3 B. Lukas, J. Peterson, J. Silver and M. T. Wilson, *Inorg. Chim. Acta, 80,* 245 (1983).
4 J. Silver, B. Lukas and G. Al-Jaff, *Inorg. Chim. Acta, 91,* 125 (1984).
5 J. Silver and B. Lukas, *Inorg. Chim. Acta, 92,* 259 (1984).
6 J. Silver and B. Lukas, *Inorg. Chim. Acta, 91,* 279 (1984).
7 J. Silver and B. Lukas, *Inorg. Chim, Acta, 106,* 7 (1985).
8 B. Lukas and J. Silver, *Inorg. Chim. Acta, 106,* 219 (1985).
9 W. I. White, in D. Dolphin (ed.), 'The Porphyrins', Vol. 7, Academic Press, London, 1978, pp 303–339 and refs. therein.
10 J. P. Collman, J. L. Hoard, N. Kim, G. Lang and C. A. Reed, *J. Am. Chem. Soc., 97,* 2676 (1975).
11 E. B. Fleisher, C. K. Miller and L. E. Webb, *J. Am. Chem. Soc., 86,* 2342 (1964).
12 M. Y. Hamed, R. C. Hider and J. Silver, *Inorg. Chim. Acta, 66,* 13 (1982).