

Concerning *bay* Salt and *peri* Chelate Formation of Hydroxyphenanthroperylene Quinones (Fringelites)

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Summary. The bathochromic shifts in the diffuse reflectance UV/Vis spectra of certain fringelite containing fossil species and the exceptional chemical stability of the fringelites and their resistance against leaching on a geological time scale can be understood from the unique complexation behavior of the fringelites with transition metal ions. According to an absorption spectroscopic study of the model system fringelite D-alkaline earth metal and transition metal ions, the fringelites are able to form *peri* chelate complexes. In addition, fringelites bearing *bay* hydroxyl groups are able to form polymeric phenolates with transition metal ions as well as with alkaline earth metal ions. This behavior leads to a complex network lattice consisting of these polymeric chains crosslinked *via* chelate coordination at the *peri* regions to transition metal ions like iron.

Keywords. Alkaline earth metal ions; Deprotonation; UV/Vis spectra; Fossil fringelites.

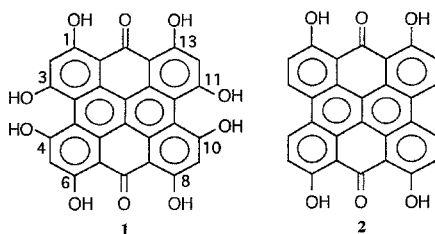
Zur Salz- und Chelatbildung bei Hydroxyphenanthroperylenchinonen (Fringeliten)

Zusammenfassung. Die bathochromen Verschiebungen in den diffusen Reflexionsspektren gewisser Fringelit enthaltender Fossilien sowie die ungewöhnliche chemische Stabilität der Fringelite und deren Widerstandsfähigkeit gegen Auswaschung innerhalb geologischer Zeiträume können aus dem ausgeprägten Komplexierungsverhalten der Fringelite mit Übergangsmetallionen verstanden werden. Aus der absorptionsspektroskopischen Studie des Modellsystems Fringelit D-Erdalkali- und Übergangsmetallionen wird abgeleitet, daß die Fringelite fähig sind, *peri*-Chelate zu bilden. Zusätzlich dazu können Fringelite, die über *bay*-Hydroxylgruppen verfügen, polymere Salze mit Erdalkali- und Übergangsmetallionen bilden. Dies führt zu einem komplexen Gitterwerk, das aus diesen polymeren Ketten, welche durch die Chelierung mit Übergangsmetallionen im Bereich der *peri*-Hydroxylgruppen vernetzt sind, besteht.

Introduction

Hydroxyphenanthroperylene quinones include a variety of naturally occurring compounds, like hypericin [1] and stentorin [2] which even exhibit interesting physiological activities. Among them, the fringelites are an exceptionally interesting type of compounds as they occur in fossils several hundred millions of years old [3, 4]. The occurrence of these pigments has been investigated using a diffuse reflectance spectroscopic method [5]. Besides the most extensively hydroxylated

fringelite D (**1**), there exists a series of fringelites which differ from **1** by the number of hydroxylic groups; fringelite H (**2**) is the member with the lowest hydroxylic group content.



Although the chemistry of phenanthroperylene quinones, and in particular that of **1**, has been investigated to some detail [6–8], it is not yet settled if these materials occur in fossil specimens in a free or bonded state. In principle, there are two possibilities for the fringelites to interact with inorganic rock materials: either to form phenolates with their extremely acidic *bay* hydroxyl groups (positions 3, 4, 10, 11) or to establish chelate type coordination complexes involving the *peri* hydroxyl and oxo groups (positions 1, 6, 8, 13, and 7, 14). This question has been brought to our attention by the observation that certain fossil species displayed significantly red shifted fringelite absorption spectra which could result from the formation of coordination compounds. Moreover, an answer could also contribute to the understanding of the problem of the incredible chemical stability of these organic chemofossils. Accordingly, the question of salt formation *versus* chelate type complexation of the fringelites will be addressed in this paper.

Results and Discussion

Fringelite D (**1**) was thought to serve as a well suited example for an investigation of the conditions of salt and chelate formation of fringelites. Moreover, due to a rational synthesis **1** was made easily accessible [7]. This compound has been found to exhibit a series of deprotonation steps [7, 9]. For the first two narrowly spaced steps pertaining to the deprotonation of the two *bay* regions (positions 3 and 10), pK_a values (20% dimethylsulfoxide/water) of 1.4 and 3.3 have been advanced. Thus, **1** constitutes a rather strong dibasic acid which is about three orders of magnitude stronger than common carboxylic acids. Deprotonation at the *peri* hydroxylic groups (positions 1, 6, 8, 13) needs a strong base: the first of these protons is characterized by a pK_a value of 12.4. Accordingly, one could infer that **1** would easily form *bay* diphenolates even with weak bases. Indeed, it has been shown that hydroxyphenanthroperylene quinones can be easily derivatized into salts with monovalent bases, and that hypericin occurs in its main natural source, *Hypericum perforatum*, as its potassium salt [10]. This salt formation can be easily monitored from the changes in chromophore absorption. Figure 1 displays the typical UV/Vis spectra observed upon stepwise deprotonation of **1** with one to three equivalents of tetrabutylammonium hydroxide. Whereas the *bay* phenolates were characterized by strong and structured bathochromically shifted absorption bands, additional deprotonation at the *peri* position led to rather broad and unstructured absorptions.

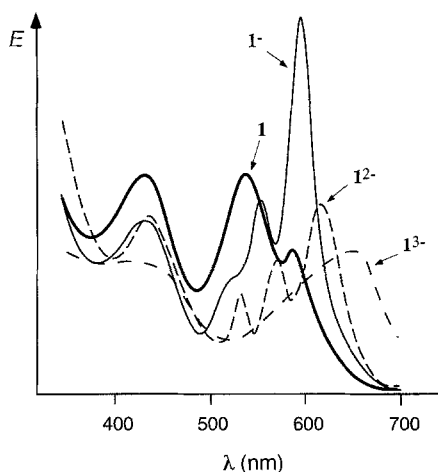
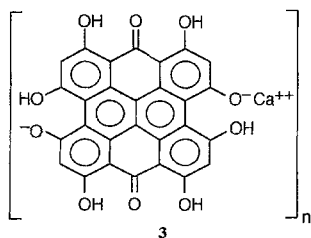


Fig. 1. UV/Vis spectra (arbitrary units) of **1**, **1**⁻, **1**²⁻, and **1**³⁻ in 80% dimethylsulfoxide/water with tetrabutylammonium as counterion

Spectrophotometric titration of **1** with one mol of weak divalent bases like $\text{Mg}(\text{OH})_2$ or $\text{Ca}(\text{OH})_2$ in 80% dimethylsulfoxide/water or in methanol produced exactly the spectrum of **1**²⁻ as displayed in Fig. 1. From these data one could envisage a polymer consisting of **1**²⁻ linked *via bay* salt bonds to the alkaline earth metal ion as given below for the calcium derivative (**3**). It should be mentioned that solutions of the Ca^{++} or Mg^{++} salts of **1** are rather unstable due to the rapid precipitation of these extremely insoluble products. Attempts to isolate and characterize these materials were complicated by coprecipitation of reactands and solvents. Nevertheless, the brutto formula could be confirmed. This behavior strongly pointed to the presence of these polymeric salts in fossil material, and is obviously the reason for the incredible stability of the pigments against leaching from fossils. Thus, according to *Blumer's* [3] and our own observations, the fringelite pigments could not be extracted directly from such species but needed treatment with strong acid beforehand. However, a third deprotonation step in **1** could not be detected upon addition of further Mg^{++} or Ca^{++} equivalents. This result was also obtained when *Hünig's* base (N-ethyl-N,N-diisopropylamine) was added in 1-3 equivalents before addition of the divalent base. The latter experiment also indicated that addition of three equivalents of *Hünig's* base was not sufficient to remove more than two protons from **1**. This is in accordance with the finding that fringelite H (**2**), which lacks the *bay* hydroxyl groups, is not deprotonated by *Hünig's* base.



To study the transition metal ion interactions of fringelites, zinc was selected as an example. In contrast to iron or copper, the interference of the ligand field

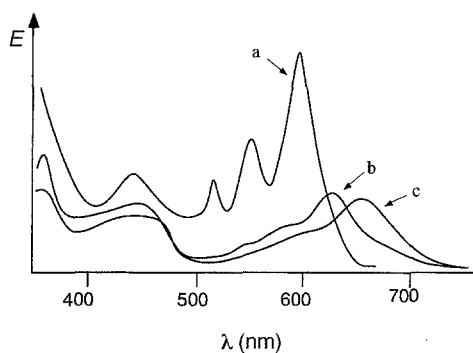
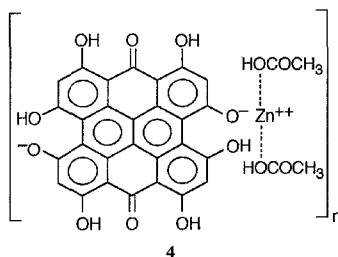


Fig. 2. UV/Vis spectra (arbitrary units) of 1^{2-} (a) and its zinc acetate titration spectra after addition of 2 (b) and 3 (c) mol equivalents in presence of an excess of *Hünig's* base in methanol

transitions of the cation with the absorptions of the chromophore could thus be avoided. Titration of **1** with one atom equivalent of zinc acetate in acetone or methanol led to a spectroscopic species which was characterized by a spectrum superimposable on that of 1^{2-} of Fig. 2. Thus, in analogy to the alkaline earth metal ions, a *bay* salt of type **4**, but additionally stabilized from interactions with the resulting acetic acid, was formed. Attempts to isolate and characterize **4** partly failed due to the varying water and acetate contents and coprecipitation of unreacted ligand in the extremely insoluble product which could not be properly controlled. Nevertheless, zinc, hydrogen, nitrogen, and carbon elemental analyses resulted in values close to the expected ones for $1^{2-}\text{-Zn}^{2+}(\text{CH}_3\text{COOH})_2$.



However, in contrast to the alkaline earth metal ions discussed above, titration of **1** in presence of an excess of *Hünig's* base with zinc acetate indicated formation of two discrete spectroscopic species at two and three mole equivalents zinc ions as illustrated in Fig. 2b. A *Job* plot (Fig. 3) corroborated the 1:2 stoichiometry of the first species.

With respect to the nature of this species it was obvious that it was different from the polymeric 1:1 salt formed without addition of *Hünig's* base. From its spectroscopic features it could be described as a species deprotonated at the *bay* regions as well as at one *peri* hydroxyl group (compare Fig. 1). It is therefore concluded to contain the structural features illustrated in formula **5**. Accordingly, a polymeric chain comparable to those of **3** and **4** accommodate an additional *peri*

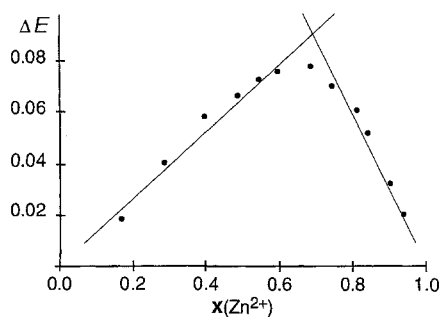
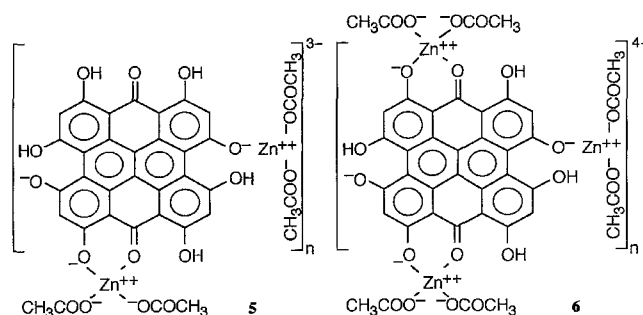


Fig. 3. Job plot for the 1-zinc(II)-complex (1:2) in methanol and an excess of *Hünig's* base, monitored at 634 nm

chelated zinc ion stabilized by coordination to two acetate ligands, with *Hünig's* base providing the necessary counter ions.



Derivatives of the above nature could be isolated. However, their elemental analysis data, although close to the calculated ones for **5**, scattered rather broadly due to varying water, acetate, and coprecipitating reagents in the extremely insoluble materials.

Addition of a further equivalent of zinc ions as shown in Fig. 2c led to an additional red shift of the absorption spectrum indicating loss of a further *peri* hydroxyl proton and, accordingly, formation of a polymeric *bis-peri* chelate type material as shown in formula **6**.

The results reported above for the zinc ion compared favorably with those obtained for other transition metal ions like iron, nickel, and copper. However, due to the spectral overlap of ligand and ligand field transitions and complications from hydrolysis and solvolysis of these ions, the spectra were less conclusive. Nevertheless, the general features, *i.e.* to form *peri* chelates in addition to polymeric *bay* phenolates, was clearly discernible. These derivatives were characterized similarly to the zinc coordination compounds **5** and **6** by structureless and bathochromic shifted absorption spectra. It was also interesting to note that with fringelite H (**2**), similar spectroscopic features were obtained, although much less pronounced. Obviously, the concomitant *bay* deprotonation in **1** played a critical role in enhancing the degree of spectroscopic changes between the different species.

According to these results, the bathochromically shifted diffuse reflectance UV/Vis spectra [5] observed for certain fossil species could be easily interpreted as stemming from a chelate *peri* complexation of the fringelites with transition metal

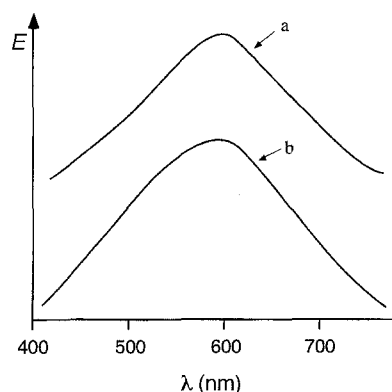


Fig. 4. Diffuse reflectance UV/Vis spectra of the Fe^{3+} precipitate of **1** applied to a TLC plate (a) and of *Spinocyrtia ostiolata* SCHLOTHEIM (b)

ions like iron which are rather abundant in certain fossil sites. This hypothesis could be also corroborated by Fig. 4, in which an iron(III) precipitate of **1** was applied to a TLC plate; the diffuse reflectance spectrum of this sample was compared to that obtained from the Devonian brachiopod *Spinocyrtia ostiolata* SCHLOTHEIM [4]. The two spectroscopic traces were found to be quite similar.

Conclusions

The bathochromic shifts in the diffuse reflectance UV/Vis spectra of certain fringelite containing fossil species can be understood from the unique complexation behavior of the fringelites with transition metal ions, especially those of iron. According to the studies reported above, they are able to form *peri* chelate complexes. In addition, fringelites substituted with *bay* hydroxyl groups form polymeric phenolates with transition metal ions as well as with alkaline earth metal ions. The probability that sufficiently basic ligands saturate the coordination of *peri* bonded transition metal ions as derived for **5** and **6** is high; therefore, a complex network lattice involving polymeric chains as illustrated in Fig. 5 would be

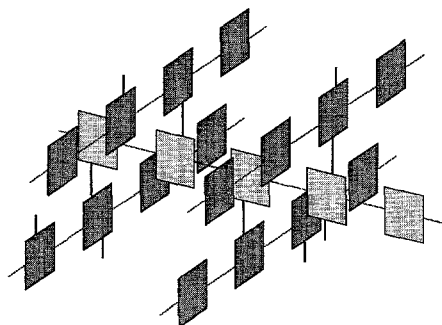


Fig. 5. Schematic structural aspects of the polymeric fringelite complex lattice in the mineral matrix of fossils; squares denote fringelites D, dashed lines connecting them notify *bay* phenolate bonds to alkaline earth metal or transition metal ions, and solid lines denote links of the thus chain forming fringelites *via* coordination to a transition metal ion

plausible. Such structural features could then nicely account for the extreme chemical stability of the fringelites and their resistance against leaching on a geological time scale [4].

Experimental

Melting points were taken by means of a Kofler hot stage microscope (Reichert, Vienna). IR and UV VIS spectra were recorded using Biorad-FT-IR-45 and Hitachi-U-3210 instruments. Fringelite D (**1**) and fringelite H (**2**) were prepared according to Refs. [7] and [11]. Elemental analyses (C, H, N) were provided by the *Mikroanalytisches Laboratorium* of the University of Vienna, and the atomic absorption analyses (Mg, Ca, Zn) were recorded using a Perkin Elmer 460 AAS instrument. The *Job* plot was recorded using standard procedures [12]. Solvents and metal salts were of analytical quality.

Fringelite D-calcium (1:1) complex (3; C₂₈H₁₂O₁₀Ca)

20 mg **1** were dissolved in 200 ml acetone and titrated under vigorous stirring with a 1% aqueous solution of Ca(OH)₂ until 1 atom equivalent of Ca was added at room temperature. After stirring for two hours at room temperature, the precipitate was centrifuged off, washed several times with distilled water, and dried in high vacuum.

Yield: 20 mg; m.p.: > 340°C; UV/Vis (DMSO): λ_{max} = 596 (25000), 550 (12000) 516 (4000) 433 (9000) nm (ε); IR (KBr): ν = 1600, 1580, 1450, 1400 cm⁻¹; the C and H elemental analyses gave best fit with 1·Ca.

Fringelite D-zinc (1:1) complex (4; C₃₂H₁₈O₁₄Zn)

20 mg **1** were dissolved in 200 ml acetone. Under vigorous stirring, 18 mg Zn(CH₃COO)₂·2H₂O were added at room temperature. After stirring for two hours at room temperature the precipitate was centrifuged off, washed several times with distilled water, and dried in high vacuum.

Yield: 16 mg; m.p.: > 340°C; UV/Vis (DMSO): λ_{max} = 596 (25000), 550 (12000) 516 (4000) 433 (9000) nm (ε); IR (KBr): ν = 1600, 1580, 1450, 1400 cm⁻¹; the C, H, and Zn elemental analyses gave best fit with 1·Zn(CH₃COO)₂.

Fringelite D-zinc (1:2) complex (5; C₃₆H₂₄O₁₈Zn₂)

20 mg **1** were dissolved in 200 ml acetone. 200 mg N-ethyl-N,N-diisopropylamine and subsequently 36 mg Zn(CH₃COO)₂·2H₂O were added at room temperature under vigorous stirring. After stirring for two hours at room temperature the precipitate was centrifuged off, washed several times with distilled water, and dried in high vacuum.

Yield: 18 mg; m.p.: > 340°C; UV/Vis (DMSO): λ_{max} = 630 (10000), 450 (8000) nm (ε); IR (KBr): ν = 1580, 1440, 1400 cm⁻¹; the C, H, N, and Zn elemental analyses gave best fit with 1·Zn₂(CH₃COO)₄.

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- [4] Falk H (1997) Fossilien (in press). It should be mentioned that for the following fossils the occurrence of fringelites could be established using the method described in Ref. [5]: *Millerocrinus munsterianus* (ORBIGNY), Upper Jurassic/Oxford, Liesberg, Baselland, CH; *Solenopora jurassica* (BROWN), Upper Jurassic/Oxford, Novion-Porcien, Dép. Ardennes, F; non determined stromatolite, Lower Triassic/Buntsandstein, Huy, NW of Halberstadt, Harzvorland/Sachsen-Anhalt, D; *Encrinus robustus* (ASSMANN) or *acculeatus* (MEYER), Triassic/Muschelkalk, Deuna/Worbis, Thüringer Becken, D; *Encrinus* cf. *brahli* (OVERWEG), Triassic/Unterer Muschelkalk, Weißenborn/Göttingen, D; *Chelocrinus carnalli* (Beyrich), Triassic/Muschelkalk, Elvise/Göttingen, D; *Encrinus liliiformis* (LAMARCK), Triassic/Muschelkalk, Alverdisen, D; *Bacterocrinus fusiformis* (ROEMER), middle Devonian/Eifel-Givet, Loogh layer, Weinberg/Kerpen, Eifel, D; *Cupressocrinites oprassus* (GOLDFUSS), middle Devonian/Eifel-Givet, Loogh layer, Meerbüsch, Hillesheimer Mulde, Eifel, D; *Eucalyptocrinites* sp., middle Devonian/Eifel-Givet, Loogh layer, Meerbüsch, Hillesheimer Mulde, Eifel, D; *Spinocyrtia ostiolata* (SCHLOTHEIM), middle Devonian/Eifel, Junkerberg layer, Gondelsheim, Prümer Mulde, Eifel, D. It should also be mentioned that occurrence of fringelite containing fossils seems to be limited to non-Alpine sites of Europe. Obviously, the more quiet sediments of these areas allowed for the preservation of these chemofossils, whereas the higher temperatures associated with the formation of the Alps destroyed them (a screening of Alpine fossils of Devon to Paleocene age from Austria provided hitherto no indication of the occurrence of fringelites).
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