

27. TETRAPYRROLE PIGMENTS FROM BERMUDA RISE: DSDP LEG 43

S. E. Palmer,¹ W. Y. Huang,² and E. W. Baker,³ Northeast Louisiana University, Department of Chemistry, Monroe, Louisiana

INTRODUCTION

Coring during four recently completed DSDP legs in the Atlantic, including the one reported here, has recovered Lower and Upper Cretaceous sediments rich in organic carbon. Geochemical studies of samples from these legs have confirmed and extended ideas concerning the diagenesis of chlorophyll in marine environments.

The transition from free-base porphyrins to metalloporphyrins (nickel and vanadyl) was clearly illustrated in Lower Cretaceous sediments collected during Leg 40 from the Cape and Angola basins at a depth of burial of 1000 to 1190 meters (Baker et al., 1978a). Even later diagenetic products, resembling the petroporphyrins, were present in Leg 41 Upper Cretaceous sediments (940 to 980 m) of the Cape Verde Rise (Baker et al., 1978c). The molecular weight distributions of these pigments, transalkylated nickel and vanadyl porphyrins, demonstrated the influence of relatively high temperatures imposed by igneous intrusive activity.

Intermediate diagenetic products (chlorins, free-base porphyrins, and nickel porphyrins) were found in Lower to Upper Cretaceous sediments collected during Leg 41 at 440 to 880 meters, and during Leg 44 in the Lower Cretaceous of the Blake-Bahama Basin at 690 to 960 meters (Baker et al., 1978c, 1978d).

Analysis of each of the above suites of Cretaceous sediments resulted in abundant data concerning a rather specific state of chlorophyll diagenesis. In contrast to these previous studies, extracts of Leg 43 samples contained intermediate diagenetic products and porphyrins distinctly representative of a later stage, that of dealkylation. In this study, Cretaceous sediment samples from Site 387 were found to contain primarily chlorins and nickel porphyrins not unlike those found in Leg 44 Cretaceous black shales and may represent samples of the same shale unit. However, late diagenetic products, dealkylated nickel porphyrins, were recovered from black shales in Site 386. One sample, Core 386-43, contained free-base and vanadyl porphyrins in addition to chlorins and nickel porphyrins. The youngest samples (Pleistocene to Miocene) contained only chlorins, while the most deeply buried Cretaceous sediments contained only nickel porphyrins. Data are presented in Table 1.

¹ Present Address: Florida Atlantic University, Dept. of Geology, Boca Raton, Florida.

² University of New Orleans, Biol. Sci. Dept., New Orleans, Louisiana.

³ Florida Atlantic University, College of Science, Boca Raton, Florida.

EXPERIMENTAL

The 16 core samples analyzed (100 grams each) in this study were stored frozen. Mixtures of acetone:methanol (9:1) were used for ball mill extraction of pigments. Crude extracts containing sufficient amounts of pigment (1 μg or more) were chromatographed over Sephadex LH-20 with THF. (Several crude extracts had one μg , or less total pigment, typically chlorin plus nickel porphyrin; the pigments in these samples were separated by HCl-ether partition). Chlorins and free-base porphyrins were isolated from the crude extract and purified by ether-HCl partition after diazomethane derivatization. The metalloporphyrins, which remained in the ether partition layer, were thus separated from the chlorins and free-base porphyrins. The nickel and vanadyl porphyrins were then separated and purified by chromatography over Grade III Alumina with cyclohexane:benzene (50:50) and benzene:dichloroethane (50:50), respectively.

Each step of sample work-up was monitored by UV-visible spectrophotometry using a Beckman Acta CIII scanning spectrophotometer. Mass spectra of porphyrins were recorded using the solid probe injection system of a DuPont 491-BR mass spectrometer. The source temperature was held at 275°C to 285°C while the probe temperature was increased in 5° increments, expelling the more volatile nonporphyrin compounds, until the desired probe temperature was reached (generally 240°C). At this point, temperature was held constant and the mass spectrum recorded. Some of the nickel porphyrin isolates which did not have a sufficiently high degree of volatility required use of the inverted probe tip method (initially described by Baker, Palmer, and Parrish, 1976).

RESULTS

Distribution With Respect to Depth of Burial and Age

Age and depth of burial are important factors in the distribution of pigments isolated from Leg 43 cores. The shallow, geologically younger sediments of Site 382 contained only early diagenetic chlorins. Site 387 sediments, collected at 263 to 787 meters, range from Eocene through Lower Cretaceous. Small amounts of chlorins (0.004 to 0.55 $\mu\text{g}/\text{g}$) and nickel porphyrins (0.01 to 0.17 $\mu\text{g}/\text{g}$), representative of an intermediate diagenetic stage, were found in all samples from this site.

The deepest core samples in this study are from Site 386 (Core Sections 60-5 and 63-1) and were collected

TABLE 1
Sample and Pigment Data Obtained From DSDP Leg 43 Cores

Section	Geologic Age ^a	Lithologic Description	Depth of Burial ^a (m)	Organic Carbon ^a (% by wt.)	Individual Yield ^b (ug/g)	UV-Visible Absorption Spectra (nanometers)					Pigment Type	
382-1-4	Pleistocene	Calc. brown mud	56.5	0.3	0.42	412	500	534	608	668	Chlorin	
382-6-4	Pliocene/Miocene	Green-gray mud	233.8	0.1	0.01	394			(590)	663	Chlorin	
382-7-0	Late Miocene	Green-gray mud	269.1	0.4	0.003	393				662	Chlorin	
386-43-3	Cenomanian	Dark gray shale	740.6	7.7	12.82	404	500		595	642	Chlorin	
	Late Cretaceous									659		
					6.91	402	500	531	571	619	Free-base porphyrin	
					2.23	400		523	552	(595)	Nickel porphyrin	
					4.87	410		533	572		Vanadyl porphyrin	
386-44-3	Cenomanian	Blue-green shale	750.1	1.9	none							
386-45-0	Cenomanian	Calc. black shale	764.8	0.5	0.01	398		522	552	(590)	Nickel porphyrin	
386-56-3	Albian	Calc. gray shale	873.6	0.9	0.15	391		512	549		Nickel porphyrin	
	(Early Cretaceous)											
386-57-3	Albian	Dark gray shale	883.2	0.6	Tr	(395)			(550)		Nickel porphyrin	
386-60-5	Albian/Aptian	Calc. dark gray	914.5	2.4	10.99	394		515	550		Nickel porphyrin	
386-63-1	Albian/Aptian	Black shale	937.1	4.0	2.42	393		514	550		Nickel porphyrin	
387-16-2	Eocene	Gray lime mud	262.9	0.3	0.03	408			(590)	661	Chlorin	
					0.01	395		517	551		Nickel porphyrin	
387-26-2	Danian	Calc. dark gray shale	414.9	0.6	0.004						658	Chlorin
	(Late Cretaceous)				0.01	395		515	551	(590)	658	Nickel porphyrin
387-35-3	Albian	Black shale	550.	2.3	0.55						658	Chlorin
					0.10	396		517	551		657	Nickel porphyrin
387-36-2	Albian	Black, gray-blue laminated shale	558.1	2.9	0.004						657	Chlorin
					Tr	395		515	551	(595)	641	Nickel porphyrin
387-37-2	Aptian	Unbedded black mud	577	3.0	0.11						641	Chlorin
					0.17	396		518	551		659	Nickel porphyrin
387-49-4	Berriasian-Valanginian	Finely laminated gray limestone	786.9	0.6	0.01						659	Chlorin
	(Early Cretaceous)				0.04	395		514	550		659	Nickel porphyrin

^aData obtained from Leg 43 Summary

^bIndividual pigment yields calculated by using the following molar extinction coefficients: pheophytin "a" = 63700 at 660 nm, free base porphyrin = 6540 at 618 nm, nickel porphyrin = 34820 at 550 nm, and vanadyl porphyrin = 26140 at 570 nm.

between 915 and 937 meters below the sea floor. Relatively high yields of nickel porphyrins were obtained. As discussed later, these particular pigments are mass spectrometrically similar to those recovered from previously studied Cretaceous sediments but are markedly different from those collected at shallower depths.

In the Upper Cretaceous sediment of Core 386-43, a large amount of extractable organic matter, corresponding to high organic carbon content (7.69%), was present. As could be expected, this sample, collected at 741 meters depth, contained chlorins and nickel porphyrins similar to those found in Site 387 sediments.

Also present were free-base and vanadyl porphyrins. The occurrence of these classes of pigments together is surprising. Free-base porphyrins have previously been found in association with chlorins such as those reported here. Vanadyl porphyrins have been found associated with nickel porphyrins and free-base porphyrins, but do not generally occur with chlorins (see Discussion). Ten and twenty-five meters below Core 386-43, pigment concentrations drop to 0.01 $\mu\text{g/g}$ of sediment. Changes in pigment concentrations and characteristics in these cores (386-43,-44,-45) may be influenced by the inconsistencies in lithology.

Characterization of Chlorins

The two methods used for isolation of chlorin fractions (Sephadex LH-20 chromatography and HCl-ether partition) also have qualitative value. Chlorins from the Pleistocene of core Section 382-1-4 were isolated in an early-running fraction from Sephadex LH-20 and extracted into 20 percent HCl. These two data, coupled with the UV-visible spectrum, are indicative of pheophytin-like chlorins. In contrast, the chlorin

fraction isolated from Upper Cretaceous Core 386-43 co-chromatographed with the free-base and metallo-porphyrins and was extractable into 10 percent HCl. In the UV-visible spectrum, the red band was shifted to lower wavelengths (657-659 nm) for all the Cretaceous chlorin samples.

Increased purity of chlorin fractions is generally achieved through multiple HCl extractions. When this method was applied to the chlorin fraction from Core 386-43, changes in the UV-visible spectrum were observed. The red band initially appeared at 659 nm; as isolation progressed, a second red band appeared at 642 nm. After completion of the third HCl extraction, the 642 nm band predominated and peaks at 500 and 619 nm, representative of free-base porphyrin, appeared. In addition, the Soret band narrowed and moved from 410 to 403 nm. These changes in the spectrum, coupled with the low HCl number, suggest that chlorins of this type are diagenetically very close to the chlorin-porphyrin transition. Ease of artifact formation makes study of this particular chlorin fraction difficult.

Mass Spectrometric Analyses of Porphyrins

Free-base porphyrins: The molecular weight distribution for free-base porphyrins isolated from Core 386-43 were similar to those reported previously (Baker et al., 1977a, c). These data are reported in Table 2. Five members of a DPEP homologous series were observed, with m/e 476, the nominal molecular weight of DPEP, predominating. Small mass spectral peaks were also observed at m/e 478, 450, 436, and 422. Pending further study, these may be free-base porphyrins of the etio or phyllo series (both being 310

TABLE 2
Mass Spectrometric Data of Free Base and Metalloporphyrins From Leg 43 Sites 386 and 387

	Carbon Number											
	33	32	31	30	29	28	27	26	25	24		
386-43												
Free-base porphyrin		476	462	449	434	420						DPEP series
		100	23	15	7	3						
Nickel porphyrin		537	523	509								Unidentified series
		100	47	25								
		539	525	511	497	483						Unidentified series
		15	10	16	22	22						
		532	518	504	490	476						DPEP series
		28	28	38	31	22						
		534	520	506	492	478	464	450				etio series
		5	5	11	16	23	28	50				
Vanadyl porphyrin		541	527	513	499							DPEP series
		87	100	70	43							
386-60												
Nickel porphyrin	546	532	518	504	490	476	462	448	434			DPEP series
	20	51	50	59	43	30	18	10	4			
	548	534	520	506	492	478	464	450	436			etio series
	2	16	72	100	96	81	41	23	9			
386-63												
Nickel porphyrin	546	532	518	504	490	476	462	448	434			DPEP series
	20	51	50	59	43	30	18	10	4			
	548	534	520	506	492	478	464	450	436	422		etio series
	2	23	41	82	85	100	66	43	21	10		
387-16												
Nickel porphyrin					497	483	469	455	441			Unidentified series
					41	81	86	100	86			
				504	490	476						DPEP series
				92	35	32						
				506	492	478	464	450	436			etio series
				16	30	82	68	73	49			
387-26												
Nickel porphyrin					497	483	469	455				Unidentified series
					39	53	45	31				
				504	490	476						DPEP series
				100	80	37						
					492	478	464	450				etio series
					19	88	78	61				
387-35												
Nickel porphyrin				511	497	483	469	455	441			Unidentified series
				50	54	79	75	79	67			
					492	478	464	450	436			etio series
					50	88	79	100	75			
387-37												
Nickel porphyrin		537	523									Unidentified series
		40	40									
		539	525	511	497	483						Unidentified series
		20	25	70	60	60						
		532	518	504	590							DPEP series
		30	40	55	40							
			520	506	492	478	464					etio series
			20	39	55	100	70					
387-49												
Nickel porphyrin		537	523	509								Unidentified series
		43	36	29								
				511								Unidentified series
				15								
		532	518	504								DPEP series
		36	41	50								
			520	506	492	478	464	450	436			etio series
			36	57	64	100	71	93	57			

Note: Data are presented as normalized peak intensities for free-base, vanadyl porphyrins, and nickel porphyrins of the DPEP series. Correction for Ni⁶⁰ isotope contribution to the etio series from corresponding DPEP series was made. This same correction procedure was used for normalization of the two unknown metalloporphyrin series.

+ 14n). In support of this, a slight divergence from the typical DPEP type visible spectrum was found: band II was shifted from 562 to 571 nm.

Vanadyl porphyrins: Four members of a homologous series of DPEP-type porphyrins were mass spectrometrically observed in the vanadyl porphyrin fraction from Core 386-43. Although previously recorded spectra (Leg 40) contained three to four DPEP homologs, the m/e 541 (molecular weight of vanadyl DPEP) always made the largest contribution. In this case, the next lower homolog, representative of a C_{31} DPEP-type porphyrin, predominated.

Nickel porphyrins: The molecular weight distributions of nickel porphyrins from Cores 386-60 and -63 form a nearly symmetrical envelope (Table 2). In both samples, the etio series predominated with DPEP/etio ratios of 0.6. Nine to ten DPEP and etio homologs were present, with C_{28} to C_{30} (m/e 478, 492, 506) porphyrins predominating. In contrast to these highly dealkylated series, the C_{32} and C_{31} members (m/e 532, 518) of the DPEP series were previously reported as being major components, resulting in a "truncated" molecular weight distribution. Only two other cases where extreme dealkylation of both series and predominance of the etio over the DPEP series have been reported. The first case was a Leg 38 Eocene sample (346-15) and the second was the thermally stressed Leg 41 Cretaceous samples (368-63-2).

Small amounts of C_{33} porphyrins of the DPEP and etio series were present. Again, such values (m/e 546 and greater), which are indicative of the addition of alkyl substituents through transalkylation, have been previously observed only as minor components of the total nickel porphyrin mixture.

Mass spectrometric data of nickel porphyrins isolated from Core 386-56 were not as easily interpretable as for 386-60 and -63, and these data are not reported in Table 2. However, a dealkylated series of etio-type porphyrins was observed, with C_{26} to C_{28} porphyrins predominating.

In marked contrast with the rather simple molecular weight distributions observed for nickel porphyrins from Cores 386-60 and -63 were those isolated from samples that also contained chlorins (386-43, 387-16, -26, -35, -36, -37, and -49). The etio series again made a major contribution to the total nickel porphyrin fraction and consisted mainly of the dealkylated homologs (C_{30} to C_{27}). Four to seven members of the etio series were present. However, the DPEP series made a smaller contribution and only three or four homologs were observed.

The complexity of the mass spectra from these nickel porphyrin samples was increased by the occurrence of two homologous series 7 and 9 mass units distant from the corresponding nickel DPEP series number. The molecular weight distributions of these series often appear to mirror the distribution of the nickel DPEP and etio series. Lower homologs, suggesting dealkylation, have the greater peak intensities.

These peculiar metalloporphyrins have not been structurally characterized, and no interpretation has

been made concerning their origin or significance. They have been observed previously in deeper core samples collected from the Black Sea and in Miocene and Cretaceous samples from Blake-Bahama Basin. Pigments with these molecular weight distributions apparently occur with chlorins and may represent a different phase of diagenesis than those of the nickel DPEP-etio series.

DISCUSSION

The data reported in this study further confirm the utility of chlorophyll degradation products as indicators of environmental parameters. For example, preservation of chlorins in sediments as old as Cretaceous is indicative of a mild thermal history. In addition, the association of vanadyl porphyrins with chlorins suggests a deviation from the expected diagenetic trend.

Depth of burial (i.e., temperature) was again shown to be a more important factor than relative geologic age in determining the degree of tetrapyrrole diagenesis. DPEP and etio nickel porphyrins were found in Albian/Aptian Cores 386-60 and -63 recovered from 915 and 937 meters depth of burial while chlorin and nickel porphyrin was present in the older but more shallow sediments of Core 387-49.

The unusual combination of early and late diagenetic products in Core 386-43 drew our attention. Although we do not fully understand the processes that lead to this combination, we can offer a few possible explanations. The association of vanadyl porphyrins with chlorins suggests that either a crude oil has entered the sediment or reworking has occurred. A third possible explanation of this unusual pigment combination is the possibility that the organic material that was deposited in Core 386-43 was different from that deposited in the other samples. In addition to the out-of-place vanadyl porphyrin was the presence of abundant high molecular weight organic material. However, Cores 386-44 and -45, 20 and 25 meters deeper, contained only small amounts of nickel porphyrin, and the high molecular weight material was not observed. In the deepest samples (Cores 386-60 and -63), where nickel porphyrins were expected and found, the amount of high molecular weight material was significantly less in comparison with Core 386-43. No vanadyl porphyrins were present in any of these deeper cores.

Thus, one particular core (386-43) stands out as being a geochemical contradiction. Combined here with diagenetic products representative of the chlorin to porphyrin transition is the unexpected presence of vanadyl porphyrins, one-third as abundant as chlorins. Because vanadyl porphyrins are so thermodynamically stable, their presence is interpreted as representing a much later stage of porphyrin diagenesis.

The significance of the high degree of dealkylation observed in the total metalloporphyrin suite is not yet apparent; however, a few inferences can be made concerning individual samples. Initial "dealkylation" reactions occur in free-base porphyrins as evidenced by

molecular weight distributions (such as for the free-base fraction, core 386-43) where m/e 476 (DPEP) is the major component and three to four lower homologs (C_{31} - C_{28}) are present in lesser quantities. This same pattern is observed after chelation with nickel or vanadyl. This phase of "dealkylation" occurs at an early to intermediate stage of diagenesis under conditions of mild thermal stress.

Symmetrical molecular weight distributions observed for nickel porphyrins isolated from deep core samples, 386-60 and -63, are reminiscent of petroporphyrins, however, these, in contrast to the typical petroporphyrin, are dealkylated, with C_{27} - C_{30} porphyrins being most abundant. In these samples and in petroporphyrins, symmetry suggests a nonselective dealkylation process. In contrast with petroporphyrins, only a minor fraction of the molecular weight distribution was composed of C_{33} (or greater) porphyrins. This finding suggests that transalkylation was minimal.

A similar explanation is offered for the altered molecular weight distribution of the vanadyl porphyrins from Core 386-43. Secondary dealkylation may have influenced the observed shift in the major peak from m/e 541 to m/e 527.

In contrast with the nearly symmetrical envelopes described above, irregular molecular weight distributions were observed for the highly nonvolatile metalloporphyrins fractions associated with chlorins. Irregularity of the envelope, produced by different trends in the individual series (i.e., one may increase towards higher molecular weight members, one may decrease, and the other may tend toward symmetry), may reflect some structural or environmental factors: (1) complexity may result from more than one set of diagenetic reactions occurring in the same time span or within different time periods (i.e., initial and secondary phases); (2) an influx of pigments with a different diagenetic history. Solution to this problem may be delayed until better characterization of the odd molecular weight series of metalloporphyrins has been made.

In this study, chlorins and nickel porphyrins were found in Upper and Lower Cretaceous sediments from the Bermuda Rise Site 387 at 415 to 787 meters. In contrast, more deeply buried (874 to 937 m) Lower Cretaceous sediments at Site 386, contained only nickel porphyrins. Paradoxically, one sample of Upper Creta-

ceous sediment (386-43) at 740 meters depth, contained a full range of chlorophyll degradation products: chlorins, free-base porphyrins, and both nickel and vanadyl porphyrins. Surprisingly, free-base porphyrins were not found in other chlorin-containing samples and vanadyl porphyrins were not found in the deeper core samples of Site 386. Chlorins were found in the younger (Pleistocene to Miocene) shallower (57 to 269 m) samples of Site 382 located on the New England Sea Mounts.

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