Pergamon

Tetrahedron Letters, Vol. 38, No. 13, pp. 2347-2350, 1997

0040-4039/97 $17.00 + 0.00

PII: S0040-4039(97)00311-0

Cyclisation, Aromatisation and Expulsion Reactions of β-Carotene during Sediment Diagenesis

Jaap S. Sinninghe Damsté[1*], Marianne Baas[1], Martin P. Koopmans[1] and Jan A.J. Geenevasen[2]

[1] Netherlands Institute for Sea Research (NIOZ), Department of Marine Biogeochemistry and Toxicology,
P.O. Box 59, 1790 AB Den Burg, Texel, The Netherlands.
[2] University of Amsterdam, Faculty of Chemistry, Department of Organic Chemistry,
Nieuwe Achtergracht 129, 1018 WS Amsterdam, The Netherlands.

Abstract: A novel diaryl isoprenoid **1** with an additional aromatic ring ($C_{30}H_{38}$), formed from the carotenoid β-carotene by cyclisation, aromatisation and expulsion reactions during sediment diagenesis, has been isolated from a 50 Ma old lacustrine sedimentary rock and identified by ^1H and ^{13}C NMR spectroscopy and mass spectrometry.

Although carotenoids are relatively labile components, they may be preserved in an altered form in ancient sediments and petroleum. For example, we recently identified in a set of sediment samples ranging in age from Miocene (6 Ma) to Ordovician (450 Ma) a suite of derivatives of the diaromatic carotenoid isorenieratene **2** presumed to be formed by cyclisation, aromatisation, and expulsion reactions and reduction of double bonds.[1-2] Since these diagenetic reactions occur with the polyene double bond system of isorenieratene, they are expected to occur also with other carotenoids during sediment diagenesis. Another widely occurring diagenetic carotenoid product is β-carotane **3**, which is derived from the ubiquitous carotenoid β-carotene **4**.[3-6] It was firstly identified in the extract of the Eocene (50 Ma) Green River shale.[3] During examination of the aromatic hydrocarbon fraction of an extract of the Green River shale by GC-MS, we encountered a suite of aromatic hydrocarbons with 30, 31 or 38 carbon atoms, tentatively identified as diaryl isoprenoids with 0-2 additional aromatic rings and with mass spectra dominated by fragment ions at m/z 119 and 120, suggesting dimethylbenzene end groups. These structures indicated the occurrence of a diagenetic reaction of carotenoids not previously encountered, i.e. the aromatisation of the 1,1,5-trimethylcyclohex-5-enyl moiety of carotenoids (including β-carotene) with concomitant loss of a methyl group. Although it was tempting to assume that C-17 and C-17' of **4** had been lost, the dominance of the m/z 120 fragment ion (formed by a McLafferty rearrangement) over the m/z 119 fragment ion (caused by β-cleavage) in the mass spectra of the diaryl isoprenoids with no additional aromatic ring indicated that the dimethylbenzene moieties contained two unsubstituted *ortho* and *para* positions[7-8].

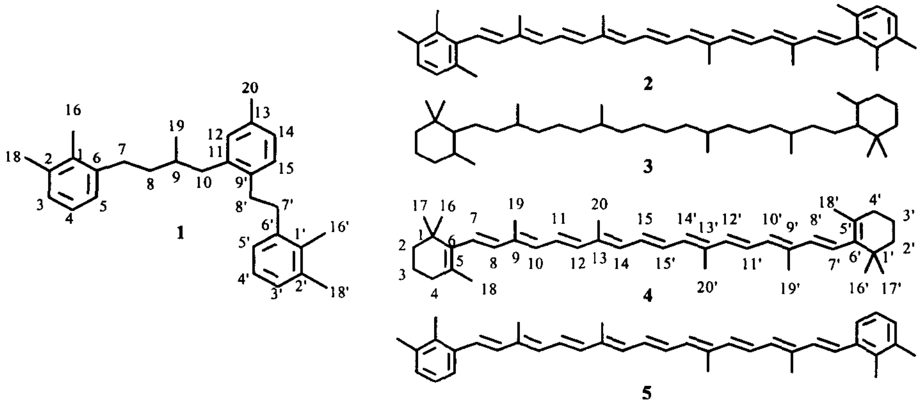

Figure 1: Structures of compounds.

To unambiguously establish the substitution pattern of the formed aromatic rings, a diaryl isoprenoid with an additional aromatic ring (1^9; $C_{30}H_{38}$) was isolated from the solvent extract of the Green River shale by column chromatography and subsequent reversed HPLC using a Polygosil C_{18} preparative column by elution with methanol/ethyl acetate (80/20; v/v). The purest fraction (1.0 mg) consisting of 74% of **1** (other 26% consisted of a complex mixture of hydrocarbons, none representing more than 10% of the total fraction) was analysed by high field ^{1}H and ^{13}C NMR (Table 1) which led to complete assignment of proton and carbon chemical shifts. Carbon multiplicities were established by APT and DEPT spectra and revealed that **1** contains 30 carbon atoms with 9 aromatic C, 9 aromatic and 1 aliphatic CH, 5 CH_2 and 6 CH_3 units. ^{1}H-^{1}H COSY and inverse one bond ^{1}H-^{13}C correlations were used to assign chemical shifts. An inverse long range (^2J and ^3J) ^{1}H-^{13}C correlation experiment (Fig. 2a) was essential for the identification of the aromatic substitution pattern of **1**. It clearly showed the connectivities of two aromatic methyl groups with only quaternary carbon atoms, establishing the position of the

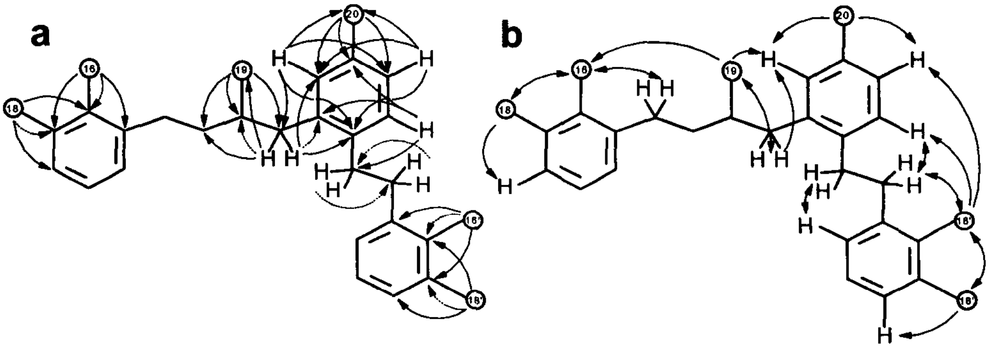

Figure 2: (a) Connectivities from an inverse long range [^2J (stippled lines) and ^3J (solid lines)] ^{1}H-^{13}C correlation experiment and (b) long-distance NOE interactions of methyl groups (C-16, C-18, C-19, C-20, C-16' and C-18'), CH_2 groups β to aromatic rings (C-7, C-10, C-8', C-7') and an aromatic CH group (C-15).

Table 1: ^1H and ^{13}C NMR data of **1**.

C-number	H-shift	C-shift[a]			
		CH_3	CH_2	CH	C
1	-				134.31[b]
2	-				136.79[c]
3	6.99 (s)			127.56	
	6.99 (s)			125.32	
5	6.99 (s)			126.79	
6	-				140.86[d]
7	2.70 (ddd, J=5, 12&13 Hz), 2.56 (ddd, J=5, 11&13 Hz)		31.80		
8	1.65 (m), 1.42 (m)		38.05		
9	1.8 (m)			34.80	
10	2.42 (dd, J=8&14 Hz), 2.69 (dd, J=6&14 Hz)		40.19		
11	-				138.83
12	6.95 (d, J=2Hz)			130.89	
13	-				135.12
14	6.97(dd, J=2&8Hz)			126.74	
15	7.11 (d, J=8Hz)			129.06	
16	2.15 (s)	14.89			
18	2.26 (s)	20.68			
19	0.98 (d)	19.55			
20	2.30 (s)	21.03			
1'	-				134.38[b]
2'	-				136.89[c]
3'	7.04 (s)			127.84	
4'	7.04 (s)			125.51	
5'	7.04 (s)			126.79	
6'	-				140.26[d]
7'	2.79 (dd; J=3&10Hz), 2.80 (dd; J=3&10Hz)		36.06		
8'	2.84 (dd; J=3&10Hz), 2.85 (dd; J=3&10Hz)		33.55		
9'	-				137.17
16'	2.24 (s)	15.01			
18'	2.29 (s)	20.72			

[a] multiplicity of signals determined by APT, DEPT(135) and DEPT(90) experiments.
[b,c,d] assignments may be interchanged.

methyl groups C-16 and C-18[10], and C-16' and C-18'[10] at C-1 and C-2, and C-1' and C-2', respectively. These assignments were confirmed by NOE experiments which revealed the interactions of the adjacent aromatic methyl groups (Fig. 2b).

The identification of **1** is the first evidence for the process of aromatisation of 1,1,5-trimethylcyclohex-5-enyl moieties of carotenoids during sediment diagenesis and has clearly established that this process does not proceed by a simple loss of one of the geminal methyl groups, but rather by a more complex and presently not completely understood reaction pathway. In fact, at present we cannot fully exclude the possibility that **1** is not derived from β-carotene **4** but from the unprecedented C_{38} diaromatic carotenoid **5**. Our earlier findings have indicated that the fossil record contains derivatives of carotenoids which have not been reported in contemporary species but may derive from carotenoids of extinct organisms.[11] This study also represents the first rigorous identification of a sedimentary carotenoid derivative formed by expulsion, in this case of *m*-xylene, from the intact carotenoid. Such a mechanism has been demonstrated to occur with intact carotenoids at elevated temperatures and during mass spectral fragmentation.[12-13] The structure of **1** also confirms the reported preferred position of the additional aromatic ring by cyclisation and aromatisation of the polyene chain of carotenoids.[1-2]

This work was supported by a PIONIER grant to JSSD from the Netherlands Organisation for Scientific Research (NWO). We thank Shell International Exploration and Production for providing a studentship to MPK and Dr. C. Erkelens (University of Leiden) for access to the 600 MHz NMR instrument. This is NIOZ Division of Marine Biogeochemistry contribution No. 440.

References and Notes

1. Koopmans, M. P.; Köster, J.; Kenig, F.; van Kaam-Peters, H. M. E.; Schouten, S.; Hartgers, W.A.; de Leeuw, J. W.; Sinninghe Damsté, J. S. *Geochim. Cosmochim. Acta*, **1996**, *60*, 4467-4496.
2. Sinninghe Damsté, J. S.; Köster, J.; Baas, M.; Koopmans, M. P.; van Kaam-Peters, H. M. E.; Geenevasen, J. A. J.; Kruk, C. *J. Chem. Soc., Chem. Commun.*, **1995**, 187-188.
3. Murphy, M. T. J.; McCormick, A.; Eglinton, G. *Science* **1967**, *157*, 1040-1042.
4. Jiang, Z. S.; Fowler, M. G. *Org. Geochem.* **1986**, *10*, 831-839
5. Carroll, A. R.; Brassell S. C.; Graham, S. A. *AAPG Bull.* **1992**, *76*, 1874-1902.
6. Vorob'yeva, N. S., Zemskova, Z. N.; Punanov, V. G.; Petrov, A. A. *Petrol. Chem.* **1993**, *33*, 277-282.
7. Kingston, E. E.; Eicholzer, J. V.; Lyndon, P.; MacLeod, J. K.; Summons, R. E. *Org. Mass Spectrom.* **1988**, *23*, 42-47
8. Sinninghe Damsté, J. S.; Kock-van Dalen, A. C.; de Leeuw, J. W. *Geochim. Cosmochim. Acta* **1988**, *52*, 2671-2677.
9. Selected mass spectral data for **1**: MS (70 eV) *m/z* 398(28), 173(75), 159(60), 119(100)
10. Note that the numbering of C-18 and C-18' in **1** does not imply any knowledge of the origins of these methyl groups.
11. Hartgers, W. A.; Sinninghe Damsté, J. S.; Koopmans, M. P.; de Leeuw, J. W. *J. Chem. Soc., Chem. Commun.*, **1993**, 1715-1716.
12. Vetter, W.; Englert, G.; Rigassi, N.; Schwietert, U. In *Carotenoids*; Isler, O., Ed.; Birkhäuser Verlag: Basel, 1971; pp. 189-266.
13. Byers, J. D.; Erdman, J. G. In *Advances in Organic Geochemistry 1981*; Bjorøy M., Ed.; Wiley: London, 1983; pp.725-732.

(*Received in UK* 3 *February* 1997; *accepted* 14 *February* 1997)