

Preserved precursors in Pumpherston shale kerogen revealed by oxidative degradation*

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(Received 6 December 2003)

Abstract: An optimised stepwise alkaline permanganate degradation was shown to have potentials in elucidating the genesis pathway and the origin of kerogens, and, more specifically, to reveal preserved precursors in a kerogen matrix. Thus, Pumpherston shale kerogen, used as the substrate in this study, was found to be of mixed origin, *i.e.*, to contain both inherited resistant algal structures (*B. braunii* A and B algaenans), as well as resistant biomacromolecular components of continental flora (cutans). It is suggested that this kerogen or parts of it were formed by the selective preservation pathway.

Keywords: kerogen, Pumpherston shale, oxidative degradation, preserved precursors.

INTRODUCTION

Kerogen, a heterogeneous, macromolecular substance of very complex composition and structure and quantitatively the most important component of sedimentary organic matter, is of biogenic origin, but the mechanism of its genesis is not yet fully explained. The Tissot-Welte “degradation-recondensation model”¹ involves diagenetic sedimentary repolymerization and recondensation reactions of biomonomers originating from the bacterial degradation of the biomass of dead auto- and/or allochthonous populations, mainly micropopulations, deposited in aquatic media. A more recent theory of kerogen genesis, the “model of selective preservation”, was formulated in 1989 by Tegelaar *et al.*² According to this mechanism, kerogen or parts of its matrix were formed by incorporation of original, highly resistant, completely preserved or slightly changed biomacromolecular tissues of precursor organisms. Since then, many investigations were aimed, on the

* Dedicated to Professor Živorad Čeković on the occasion of his 70th birthday.

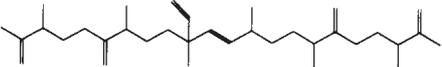
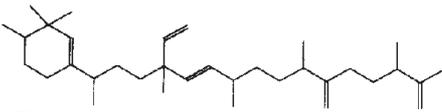
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one hand, at isolation and structure elucidation or resistant biopolymers in cell walls of algae (algaenans), bacteria (bacterans), and protective membranes of continental flora cells (cutans and suberans), and, on the other hand, at finding their unchanged fossilized forms in sediments.³⁻¹²

Among a number of different contemporary algal populations the paleoanalogs of which are considered to be possible bioprecursors of some kerogens, the first and the best studied is the green, freshwater unicellular thick cell wall microalga, *Botryococcus braunii*, i.e., its A, B and L races.¹³⁻¹⁵ According to recent data, the cell walls of this alga contain, on average, ca. 11 % of algaenan (relative to the dry sample),¹² a resistant, aliphatic biomacromolecule (Fig. 1).⁷ One of the characteristics of *B. braunii* is that it synthesizes large amounts of lipids, up to 75 wt %, some of them being extremely unusual. The composition of the lipids (involving hydrocarbons and polar lipids) is shown in Table I.¹⁶ and references therein. Due to their specific chemical and bacterial resistance, *B. braunii* algaenans represent the major part of fossilized remnants of *B. braunii* colonies in numerous sediments. In some cases the deposits of these remains are very massive, forming sediments of high oil potential, e.g., coorongite^{17,18} and torbanites.^{3,19,20}

TABLE I. Summary of lipids in two races of *B. braunii* (A and B)¹⁶

<i>B. braunii</i> race A	<i>B. braunii</i> race B
Hydrocarbons (nonpolar lipids)	
C ₂₅ -C ₃₁ odd carbon numbered <i>n</i> -alkadienes and <i>n</i> -alkatrienes, dominantly C ₂₇ , C ₂₉ , C ₃₁ 	a) C ₃₀ -C ₃₇ botryococcenes  b) Higher acyclic botryococcenes c) Cyclic botryococcenes  d) Variety of carotenoids
Polar lipids	
a) C ₁₄ -C ₃₀ even carbon numbered <i>n</i> -fatty acid triacylglycerols b) Botryals (even carbon numbered C ₅₂ -C ₆₄ α -branched and α -unsaturated aldehydes) c) <i>n</i> -Alkenylphenols d) Epoxides (epoxyalkanes, epoxybotryals and epoxyalkylphenols) e) High molecular weight lipids derived by coupling of epoxycompounds	a) C ₁₄ -C ₃₀ even numbered <i>n</i> -fatty acids b) Epoxides of polymethylated squalenes
Building blocks of resistant biopolymers	
Polymethylenic units	Polymethylenic units

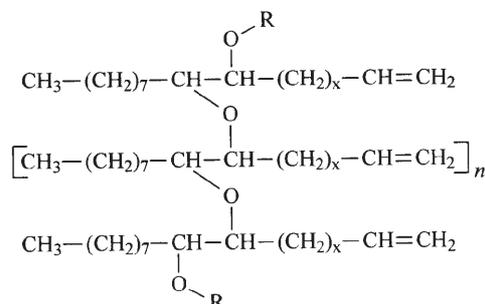


Fig. 1. The structure of algaenan of *B. braunii* A alga suggested by de Leeuw *et al.*⁷ ($n = ?$; $x = 15$, 17 or 19; $R =$ acyl groups $\text{C}_{18}\text{--}\text{C}_{30}$ of even carbon number or alkyl groups $\text{C}_{24}\text{--}\text{C}_{26}$).

Structural investigations of resistant, chemically and diagenetically stable biopolymers and parallel investigations of their fossilized forms have so far been predominantly microscopical (transmission electron microscopy) and pyrolytic (pyrolysis-gas chromatography-mass spectrometry). In this study a typical chemical degradation method, *i.e.*, optimised stepwise oxidative degradation by alkaline permanganate,²¹ was used for the structural elucidation of preserved biopolymers in a shale kerogen, in the same manner as in a recent study of Estonian kukersite kerogen.²² Namely, the results observed for the individual classes of acids, the relations between these classes, as well as their homologous series, their ranges and distributions, in the oxidation products of the Pumpherston shale kerogen matrix²³ are considered in this paper as possible indicators of fossilized resistant biomacromolecules, hence as indirect indicators of specific genesis of a corresponding part of this kerogen by selective preservation.

Pumpherston oil shale is considered to belong to the torbanites and its origin to be related to the algae *Botryococcus braunii* A and/or B and their algaenans. However, microscopic investigations of Pumpherston shale samples did not reveal the morphology characteristic of fossilized colonies of these algae.²⁴ Their organic matter was found to be microscopically heterogeneous. Some of the components could have been related to algal remnants and the rest was found to be amorphous and intimately mixed with the mineral matrix. The observed vitrinite indicated to a certain contribution of continental flora. In addition to microscopic investigations and the mentioned stepwise oxidative degradation by alkaline permanganate,²³ pyrolysis was also used for a detailed structural investigation of this shale kerogen.²⁴ It involved parallel, comparative "off-line" pyrolysis of kerogens from three pure torbanites of exclusive and unquestionable algal origin, and Pumpherston shale kerogen.

EXPERIMENTAL

Sample preparation

The Pumpherston shale sample Ca7 examined in this paper belonged to the Lower Oil Shale Group of Lothians shales (South Scotland) of Carboniferous age. It was thankfully obtained from the Newcastle Research Group in Fossil Fuels and Environmental Geochemistry, the University of

Newcastle upon Tyne. The sample contained 22.6 % organic matter. The inorganic portion was composed predominantly of carbonates (particularly siderite), containing fibrous gypsum and 1.1 % pyrite. The shale contained 0.1 % vitrinite (its reflectance being 0.59 % Ro), 0.2 % inertinite, 0.4 % sporinite, and 11.0 % *Botryococcus*.

The kerogen concentrate, prepared by the method described by Vitorović *et al.*,²⁵ contained 16.5 % ash and 82.0 % kerogen, consisting of 75.70 % C, 10.37 % H, 4.18 % N, and 9.75 % O plus S_{org} (by difference). According to the H/C and O/C ratios (1.64 and 0.10, respectively), the investigated kerogen belongs to type I kerogens.

Oxidative degradation

The kerogen concentrate (2.74 g, containing 2.25 g kerogen) was degraded in 29 steps by an optimized degradation method²¹ consisting of the stepwise oxidation of the sample by alkaline permanganate, *i.e.*, using 0.5 g KMnO₄ in 100 cm³ 1 % KOH / 2.5 g kerogen per step, at a constant temperature of 75 °C. The oxidation products were removed after each step, the products of each five consecutive steps were collected into fractions which were further separated into four types of products: neutrals and bases, ether soluble acids, ether insoluble acids (the so-called precipitated acids), and acids isolated from concentrated aqueous solutions.

The neutrals and bases were not further examined.

The precipitated acids were combined and further oxidized using the same method (0.25 g KMnO₄ per step in 55 cm³ 1 % KOH) in 31 steps at 75 °C. After reduction of each portion of the reagent, the reaction mixture was treated in the same manner as was used for the kerogen degradation products.

All soluble acids, including the acids isolated from aqueous solutions, were methylated with ethereal diazomethane, the esters were analysed by GC and identified by GC-MS.²³

RESULTS

Twenty-nine steps were necessary for the degradation of 2.25 g kerogen, and a total amount of 13.05 g KMnO₄ was consumed (5.8 g per gram of kerogen). Approximately 143 h were needed for the 28 steps. In the last, 29th step, which lasted over 30 h, the permanganate was not fully reduced, indicating the end of degradation.

The yields of oxidation products in the 29 steps are shown in Table II (A). The total recovery of the products was 83.7 %.

TABLE II. Stepwise oxidative degradation of the Pumpherson shale kerogen. Yields of oxidation products: (A) degradation of kerogen concentrate, (B) degradation of precipitated acids, (C) total yield of products

	A (% relative to kerogen)	B (% relative to precipitated acids)	C (A+B) (% relative to kerogen)
Neutrals and bases	3.7	19.2	7.9
Soluble acids	35.8	34.9	43.8
Precipitated acids	22.1		
Acids from aqueous solutions	3.3	5.4	4.5
Resistant residue	18.7	12.0	21.4
Total recovery of products	83.7	71.5	77.6
"Losses"	16.3	28.5	22.4

Thirty-one steps were necessary for almost complete degradation of 0.5 g precipitated acids, and a total amount of 7.75 g KMnO_4 was consumed (15.6 g per gram of acids). The total duration of the thirty steps was approximately 13 h. The last step lasted over 30 h. The total yields of oxidation products are shown in Table II (B). The total recovery of the products was 71.5 %.

Table II also shows the yields of various oxidation products (neutrals and bases, soluble acids, precipitated acids, and acids from aqueous solutions) and the resistant organic residue, as well as the total degradation "losses".

TABLE III. Composition of acidic oxidation products obtained by stepwise alkaline permanganate degradation of Pumpherston shale kerogen

Aliphatic acids		% Relative to total identified acids	% Relative to kerogen
<i>n</i> -Monocarboxylic	$\text{C}_9\text{-C}_{36}$	54.3	26.0
α,ω -Dicarboxylic	$\text{C}_6\text{-C}_{33}$	36.7	17.6
Branched monocarboxylic and isoprenoid acids	<i>iso</i> - C_{15} ; <i>anteiso</i> - C_{15} , $\text{C}_{17}^{\text{br}}$ C_{15}^{α} isoprenoid	0.1	0.05
Alkane polycarboxylic acids			
Tricarboxylic	$\text{C}_5\text{-C}_7$; $\text{C}_8^{(2)}$, C_9	1.4	0.6
Tetracarboxylic	C_6 , C_8 , C_9 , $\text{C}_{10}^{(2)}$, $\text{C}_{11}^{(2)}$, C_{12} , C_{13}	3.7	1.8
Aromatic acids			
Monocarboxylic		0.3	0.2
Dicarboxylic		1.6	0.8
Tricarboxylic		1.4	0.7
Tetracarboxylic		0.5	0.2

The GC-MS analyses revealed the presence of various *n*-alkane-mono- and α,ω -dicarboxylic acids, branched (*iso*- and *anteiso*-) monocarboxylic acids, alkane-tri- and tetracarboxylic acids, and aromatic mono-, di-, tri- and tetracarboxylic acids. The identified acids are listed in Table III, which also shows the proportion of the individual products, calculated relative to the total acids and relative to kerogen.

DISCUSSION

Bearing in mind the feasibilities of the employed optimised oxidative degradation method,²¹ the main aim of this study was to identify and possibly quantify the oxidation products indicative of portions of the Pumpherson shale kerogen possessing resistant biopolymer characteristics, which could thus be considered as their degradation precursors.

Normal aliphatic acids as indicators of algaenan precursors

As is well known, *B. braunii* algaenans are aliphatic biopolymers. Degradation of such structures by alkaline permanganate produces mostly aliphatic mono- and dicarboxylic acids. Their compositions, ranges and distributions are expected to reflect the character of the precursor aliphatic network, and their yield to be a reliable quantitative measure of the proportion of the corresponding aliphatic structures in the kerogen matrix. Therefore it was logical in the first instance to search for this type of oxidation products as indicators of preserved biopolymers in the examined kerogen.

n-Alkane-mono- and dicarboxylic acids were the most abundant acids in the oxidation products of Pumpherson shale kerogen: 91.0 % relative to the identified acids and 33.6 % relative to kerogen (Table III). The distribution of these acids is

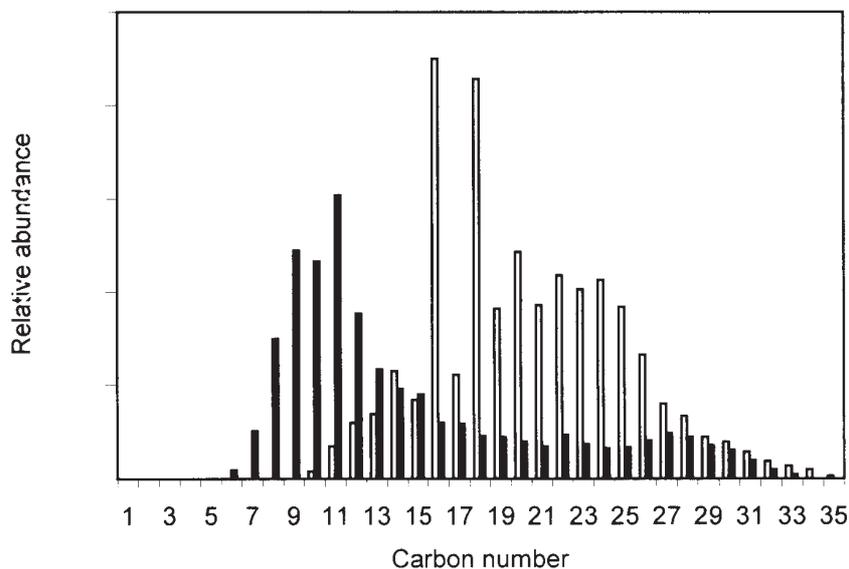


Fig. 2. Distribution of *n*-alkane-monocarboxylic (□) and α,ω -alkane-dicarboxylic (■) acids obtained by oxidation of Pumpherson shale kerogen.

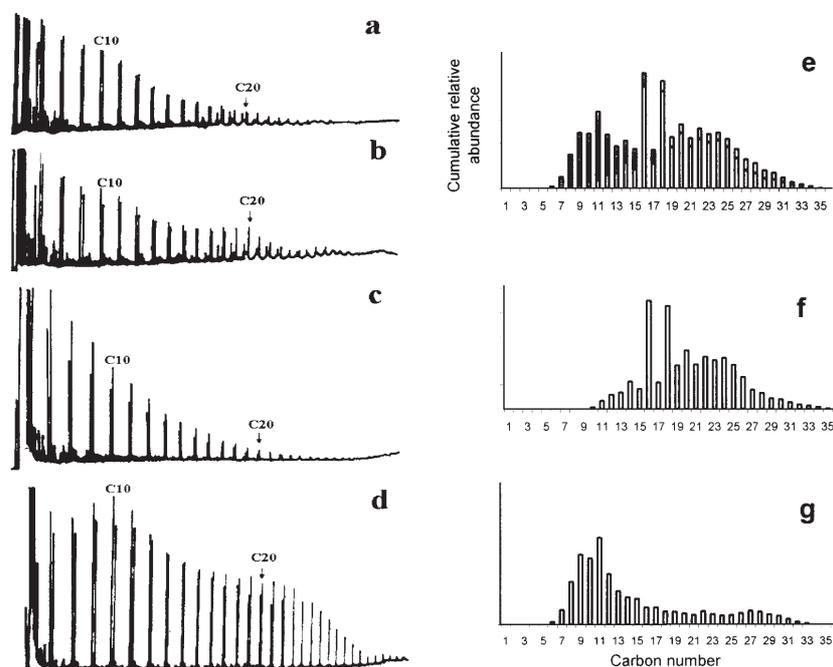


Fig. 3. Distributions of C₆₊ doublets in the low polar pyrolysates of algaenan of *B. braunii* A alga (a), algaenan of *B. braunii* B alga (b), pure torbanite (c) and Pumpherston shale kerogen (d).²⁴

Distributions of summed amounts of *n*-alkane-mono- and α,ω-alkane-dicarboxylic acids of the same carbon number (e), of *n*-alkane-monocarboxylic acids (f) and α,ω-alkane-dicarboxylic acids (g), obtained by oxidation of Pumpherston shale kerogen.

shown in Fig. 2. *n*-Alkane-monocarboxylic acids were identified in the C₉-C₃₆ range with a pronounced maximum at C₁₆ and a very similar proportion of C₁₈ acid. The range up to C₂₂ had an even/odd domination which later disappeared. α,ω-Dicarboxylic acids were identified in the C₆-C₃₅ range with a bimodal distribution, the first sharp maximum being at C₁₁ with a less pronounced sub-maximum at C₂₇. The question is whether these acids are indicators of algaenan precursors.

By comparing the alkaline permanganate degradation products with the pyrolysis products, the ranges of the identified aliphatic *n*-mono- and dicarboxylic acids were found not only to match the range of *n*-alkene/alkane doublets in the low polar pyrolysate of the same kerogen, but also to match the corresponding pyrolysates of pure *B. braunii* A and B algaenans (Fig. 3a, b and d, respectively).²⁴ Nevertheless, a quantitative comparison of the individual classes of mono- and dicarboxylic acids over the complete range is not possible since the acidic oxidation products and the pyrolysate alkene/alkane doublets do not have quite the same origin. Namely, aliphatic mono- and dicarboxylic acids, *i.e.*, the kerogen oxidation products, originate not only from alkyl substituents and bifunctionalized polymethylene chains, but also from ether and ester moieties, while alkanes and alkenes in the pyrolysates are produced by α- and β-radical scission of aliphatic

components, *i.e.*, alkyl substituents and polymethylene chains. The presence of alkene/alkane doublets in pyrolysates is generally attributed to resistant aliphatic biomacromolecules.^{26–28} The observed quantitative difference is demonstrated in Fig. 3. For the sake of comparison, the *n*-alkane mono- and dicarboxylic acids oxidation products of the same carbon number were added and are shown in the form of a histogram in Fig. 3e. By comparing the histogram 3e with the histogram 3d representing the composition of the pyrolysate of the same shale kerogen, discrepancies are particularly noticeable up to C₂₀₍₂₂₎ members of the homologous series. The distribution shown in the 3e histogram, characterized by an even/odd preference in the C₁₂–C₂₂ range and extremely high C₁₆ and C₁₈ peaks, is due mainly to *n*-monocarboxylic acids which, most probably, represent the hydrolysis products of sterically hindered ester moieties and possibly oxidation products of ether moieties as well (Fig. 1). On the other hand, a dominance of even C-numbered products was not observed in the corresponding C_{2n} and C_{2n-1} alkene/alkane pyrolytic doublets, suggesting their restricted origin in spite of the fact that some structures in *B. braunii* A derived kerogens did yield specific pyrolytic products, *i.e.*, alkadienes and alkatrienes with a C₂₇, C₂₉ and C₃₁ domination, as well as alkanones and alkenones (alk-1-en- ω -9 and alk-1-en- ω -10) also with a C₂₇, C₂₉ and C₃₁ domination.²⁹ However, the low content of the latter structures in the kerogen matrix could not explain the high yield and the specific distribution of *n*-alkane-mono-carboxylic acids up to C₂₀₍₂₂₎ in the oxidation products. Hence, it seems most probable that *n*-alkane-mono-carboxylic acids are not only oxidation products of algaenan components in the kerogen matrix, but also that they derived, to a remarkable extent, from the oxidation of hydrolysis products of ester moieties as has been clearly demonstrated for Irati or Moroccan Timahdit and Tarfaya kerogens.^{30–32} A further proof for such an assumption are the kerogen hydrolysis products, their yield, composition and distribution, *i.e.*, the observed pronounced predomination of monocarboxylic acids and their even/odd preference, low yield of C₂₀₊ homologues and sharp C₁₆ and C₁₈ maxima.

n-Alkane-dicarboxylic acids, especially in the < C₂₀ range, seem generally to be a much better indicator of the presence of algaenan structures in kerogens. For most of the kerogens examined by permanganate degradation,²¹ this narrow range always had a similar pattern and a fixed position of the maximum which depended on the algal precursor. Comparison of algaenan A and algaenan B pyrograms (Fig. 3a, b) with the histogram of alkane-dicarboxylic acids observed in the oxidation of Pumpherston shale kerogen (Fig. 3g), exhibits the same ranges in all cases, but the resemblance between the algaenan B pyrogram and the Pumpherston shale kerogen dicarboxylic acids histogram was superior. Thus, for example, an equivalent for the C₁₀ alkene/alkane doublet maximum observed in the algaenan B pyrogram (Fig. 3b) but not in the algaenan A pyrogram (Fig. 3a) was found in the Pumpherston shale kerogen histogram. However, since this doublet originates from radical scission of the corresponding polymethylenic sequences, the same source is recognized in the

alkane-dicarboxylic acids histogram as the maximum at C₁₁ (Fig. 3g). Hence, this maximum seems to be characteristic of *B. braunii* B algaenan. These facts suggest that *B. braunii* algaenan B is a constituent, at least partially, of the Pumpherston shale kerogen matrix. Another proof may be found in the slight bimodality observed in the algaenan B pyrogram and the Pumpherston shale kerogen pyrogram, with a slightly pronounced submaximum around C₂₀. The distribution of dicarboxylic acids, *i.e.*, the Pumpherston shale kerogen oxidation products, was also bimodal, with somewhat higher peaks visible around C₂₂, and a second maximum shifted to C₂₇. However, the C₂₀₊alkene/alkane doublets in the Pumpherston shale kerogen pyrolysate and C₂₀₊ dicarboxylic acids in the oxidation products of the same kerogen might be of mixed origin, *i.e.*, algaenans might not be their only origin. Besides, the origin of dicarboxylic acids is not exclusively of polymethylenic structure. For these reasons the C₂₀₊ range in the low polar pyrolysates and the same range in alkane dicarboxylic acids as oxidation products are not quite reliable indicators of algaenan precursors in kerogen matrices.

The observed general similarities between the ranges, compositions and distributions of the low polar pyrolysate of the Pumpherston shale kerogen and the α,ω -dicarboxylic acids, *i.e.*, the oxidative degradation products of the same kerogen, suggest the aliphatic skeleton of the kerogen matrix to be mostly of polymethylenic character.

Although the ranges of the identified aliphatic *n*-mono- and dicarboxylic acids were found to match the ranges of doublets in the low polar pyrolysate of pure *B. braunii* B and A algaenans, quantitative comparison of their compositions and distributions suggest the proportion of algaenan structures in the Pumpherston shale kerogen to be relatively small.

Normal aliphatic acids as indicators of cutan precursors

In addition to the above mentioned facts, it should also be added that the pyrograms of low polar pyrolysates of a pure torbanite and *B. braunii* A and B algaenans differed substantially from the Pumpherston shale kerogen pyrogram (Fig. 3a–d). The differences were illustrated by a smaller participation of *n*-alkene/*n*-alkane < C₂₀ doublets in the Pumpherston shale kerogen compared to torbanite and the corresponding algaenan pyrograms (in harmony with the participation of the corresponding carbon chains in the matrix), and by a much more pronounced participation of C₁₅₊ (particularly C₂₅₊) doublets (carbon chains in the matrix) (Fig. 3a–d). The shift in the distribution towards higher homologues is attributed to pyrolysis products of cutans, for which such ranges and distributions are characteristic.³³ This can be considered as pyrolytic proof of a possible role of continental flora (*i.e.*, cutans) in the genesis of Pumpherston shale kerogen.

On the other hand, the long chain aliphatic cutan structures in a kerogen matrix are one of the main sources of C₂₀₊ mono- and dicarboxylic acids in the oxidation products of kerogens of at least partial continental origin. The pronounced partici-

pation of both C_{20+} (particularly C_{25+}) alkyl substituents and polymethylenic chains in the Pumpherston shale kerogen led to the relatively high yields of C_{20+} mono- and dicarboxylic acids in its oxidation products (Fig. 2). This fact is probably proof of the simultaneous existence of cutans (and plant waxes) in the examined sample. Namely, (i) a smooth distribution and wide ranges (up to and above C_{30}) of both the mentioned classes of acids in the oxidation products, (ii) a particularly higher yield of C_{20+} dicarboxylic acids, (iii) their bimodal distribution, as well as a sub-maximum in the C_{20+} range (in most cases C_{27}), generally originate from cutan biopolymers, and thus indicate their presence and evidence remnants of continental flora in a kerogen matrix.

The presumption of the mixed origin of Pumpherston shale kerogen and the involvement of preserved cutan biopolymers in addition to algaenans is corroborated by comparison of its potassium permanganate oxidative degradation with the same degradation of several other typical shale kerogens (Fig. 4a, b, c, e).

Comparison with some type I, type I–II and type II kerogen degradations

For sake of comparison, Fig. 4 shows the histograms of the dicarboxylic acids obtained by degradation of kerogens from Green River,³⁴ Irati,³⁵ Aleksinac,³⁶ and Pumpherston²² (type I), S. Pyongan (type I–II),³⁷ and Whitby (type II)³⁸ shales, using the identical stepwise alkaline permanganate oxidation method.²¹

Green River shale kerogen most probably originates from the thin cell walls of fresh water algae,³⁶ with no contribution from continental flora. Just a very small resistant residue escaped the degradation of this kerogen. This means that the long polymethylenic chains of the matrix of algal origin were completely oxidized, not leaving a substantial degradation residue. However, the yields of C_{20+} acids were low.³⁴

On the other hand, the Irati shale kerogen, a purely *B. braunii* A-derived kerogen,³⁹ left a relatively large residue which was resistant towards oxidative degradation. The proportion of C_{20+} dicarboxylic acids in the oxidation products of this kerogen was also low. Bearing in mind the relatively high degradation residue, two alternative explanations of this fact are possible: (a) the corresponding long chain polymethylenic and α,ω -bifunctionalized long aliphatic chains remained preserved in the degradation residue, or (b) the degradation residue of Irati shale kerogen is not composed of such structural elements. The second presumption is more probable since it was shown that coorongite kerogen, formed exclusively by fossilization of *B. braunii* A alga residues, contained a significant proportion of polymerised alkoxy-etheral lipids which, bearing in mind the type of consolidating bonds (dominantly phenolic), should demonstrate some selective resistance towards degradation by alkaline permanganate. If this is so, the observed C_{20+} ranges of dicarboxylic acids obtained by the stepwise oxidation of Green River and Irati shale kerogens suggest that long chain polymethylenic and/or aliphatic α,ω -bifunctionalized algaenan structures in kerogen matrices may be completely oxidized, but that the share of these structures in the algaenan (and in the kerogen) is low.

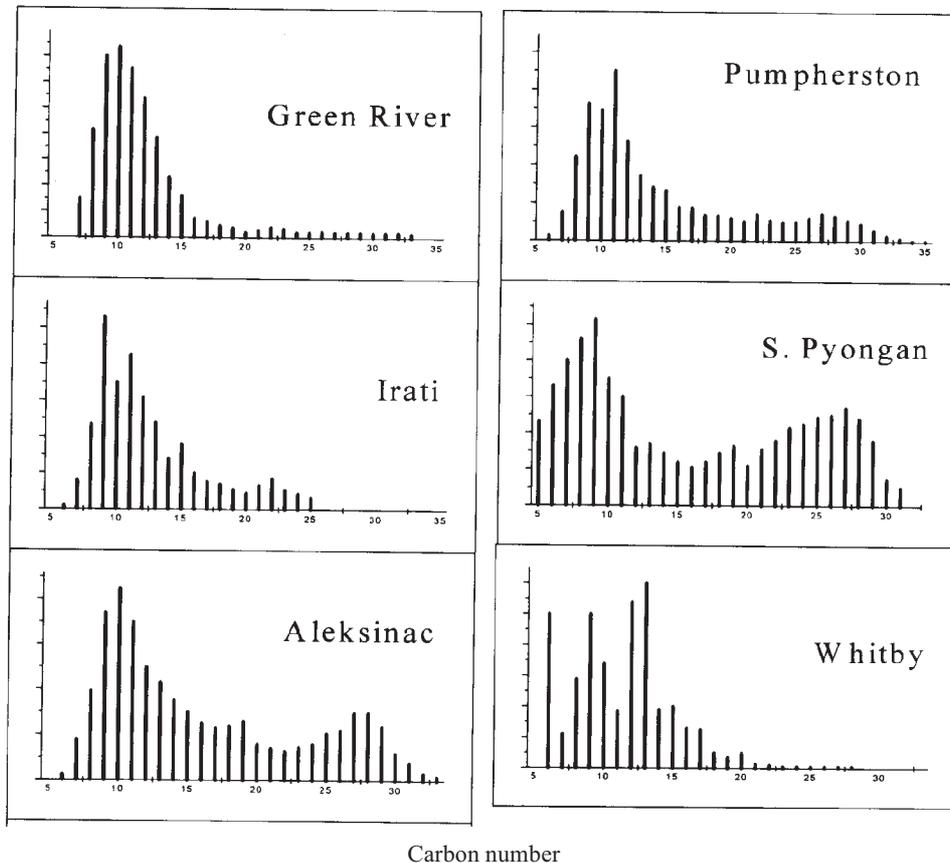


Fig. 4. Histograms of α,ω -alkane-dicarboxylic acids obtained by using an identical method of permanganate degradation of different kerogens.

Pumpherston and Aleksinac shale kerogens (Fig. 4d, c) are also of algal origin, but do not originate from the same algal populations. The sole common feature of these two kerogens is that remnants of continental flora, *i.e.*, of cutan as well, participated to some extent in their formation, and that both of them left a significant amount of residue resistant to oxidative degradation. The participation of cutan in their matrices is demonstrated by the similar characteristics of their histograms: bimodal distribution in the dicarboxylic acid homologous series, with a sub-maximum at C_{27} , and a relatively high proportion of C_{20+} acids.

The histogram of the dicarboxylic acids, observed after the oxidative degradation of the type I–II S Pyongan shale kerogen, was even more pronounced in this respect (Fig. 4e), but the degradation-resistant residue of this kerogen was relatively small. On the other hand, the histogram of the dicarboxylic acids obtained during the degradation of type II Whitby shale kerogen, which is characterized by a significant participation of continental flora in its formation, practically showed no

C₂₀₊ homologues. Moreover, the < C₂₀ of dicarboxylic acids was irregular and quite different to the same range of dicarboxylic acids obtained by degradation of pure algal or mostly alga-derived kerogens. The kerogen of Whitby shale also left a high amount of residue resistant towards degradation.

Based on these comparisons of the oxidative degradation of several structurally different kerogens, the following presumptions may be made:

- the distribution in the < C₂₀₊ range of dicarboxylic acids obtained by permanganate degradation of kerogens depends on the proportion of algaenan structures in the examined substrate, and its maximum on the kind of precursor alga;
- the C₂₀₊ range of dicarboxylic acids observed in the products of the stepwise alkaline permanganate oxidation of a kerogen is indicative of cutan precursors;
- the resistant residues observed in the degradations of kerogens of algal origin most probably are not composed of long chain aliphatic polymethylenic structures. Such algal structures, as well as cutan structures, are almost completely oxidized, and in both cases their share in the corresponding biomacromolecules and their fossilized forms are not high;
- C₂₀₊ members of “cutan” dicarboxylic acid homologous series are not detected in the oxidation products of kerogens characterized by a high contribution of flora remnants, with lignin as the dominant highly resistant precursor. This is probably due to their small amounts which are masked by the much more abundant, equally resistant lignin which does not contain such chains.

According to these presumptions, cutan is present in the matrix of Pumpherson shale kerogen and the degradation residue of this kerogen is most probably composed of some hetero-moieties, possibly containing phenoxy ethereal bonds.

Normal mono- vs. dicarboxylic acids ratios and the degradation losses

The peculiarity of the aliphatic normal mono- and dicarboxylic acids obtained by degradation of Pumpherson shale kerogen is their exceptionally high quantitative ratio: mono-/dicarboxylic = 1.48 (Table III). The existence of sedimentary aliphatic polymeric structures, or parts of these, with the major portion being in the form of alkyl substituents and ester moieties would not be quite logical. However, the high proportion of such structures, based on the oxidation products, might only be apparent. Namely, high degradation losses as well as a large resistant residue could explain this apparentness. Indeed, the oxidative degradation of Pumpherson shale kerogen left 21.4 % resistant residue and the losses were 22.4 % (in the form of CO₂ or water soluble low molecular weight mono- and dicarboxylic acids) (Table II). The high degradation losses are probably, at least to a certain degree, due to further oxidative degradation of primary oxidation products, the degradation steps of Pumpherson shale kerogen lasting exceptionally long from beginning to end. Degradation losses may also be a sign of a high proportion of short alkyl substituents in the Pumpherson shale kerogen network or, generally, of a more dense cross-linking of its polymethylene matrix.

On the other hand, the stepwise oxidation of *B. braunii* A-derived Irati shale kerogen did not produce high degradation losses but the resistant residue was high, 18.6 %. Based on the interpretation offered for the degradation of Pumpherson shale kerogen, the ratio of alkane-mono- vs. dicarboxylic acids was found to be illogically high (2.64).³⁵ An explanation of this apparent paradox could be in the fact that its oxidation products contained *ca.* 11 % aromatic acids, so that significant alkyl substitution and hence the higher yield of alkane-monocarboxylic acids could be ascribed to aromatic constituents of the matrix, as clearly demonstrated using the transalkylation reaction.^{40,41} Such an interpretation does not fit the Pumpherson shale kerogen, its oxidation products containing only 1.9 % aromatic acids (Table III). Hence, it seems that these two *Batryococcus*-derived kerogens (Pumpherson and Irati) do not contain the same aliphatic, *i.e.*, algaenan components: they differ in the proportions of short polymethylenic sequences and short alkyl chains, as well as methyl substituents.

A similar difference possibly exists between algaenans of *B. braunii* A and B algae. Namely, *B. braunii* B algaenans must have a more pronounced methyl substitution and a higher cross-linking of the polymethylene network, since they originate from isoprenoid, unsaturated botryococenes, in contrast to *B. braunii* A algaenans derived from C₂₅–C₃₃ *n*-alkadienes and alkatrienes (Table I).¹⁶ Such a presumption leads to the conclusion that part of the Pumpherson shale kerogen originates from *B. braunii* B alga in contrast to Irati shale kerogen which is *B. braunii* A-derived. Moreover, bacterial biomass with pronounced methyl substitution and shorter C-chains must have played a certain role in the formation of Pumpherson shale kerogen. This biomass contributed to the larger oxidative degradation losses, and simultaneously, to the higher yields of < C₂₀ acids. Direct qualitative proof of the role of bacterial biomass in the genesis of Pumpherson shale kerogen are the small amounts of branched acids identified in the oxidation products.

Seemingly, the presumption on higher participation of shorter alkyl chains in Pumpherson shale kerogen is not corroborated by pyrolytic experiments (Fig. 3d).²⁴ However, as mentioned, the “off-line” pyrolysis used in this case was shown to produce a loss of lower hydrocarbon homologues. On the other hand, higher yields of C₂₀₊ hydrocarbon doublets in Pumpherson shale kerogen pyrolysates, presumably originating from cutans, diminished the relative proportion of shorter chain doublets in the pyrolysis products. Nevertheless, this does not mean that shorter chains were less abundant in Pumpherson shale kerogen than in *B. braunii* A and B algaenans, or in pure torbanite, the pyrolyses of which were carried out in parallel (Fig. 3).

Alkane-polycarboxylic acids

Kerogen oxidation products generally contain different amounts of alkane-tri- and tetracarboxylic acids. These acids are considered to be a good quantitative indicator of alicyclic and heterocyclic structural elements in a kerogen matrix, but they cannot serve as a basis for differentiation of their precursors in the matrix. By

pyrolysis, these structures are converted into different substituted cycloalkanes, cyclic sulphur and oxygen compounds, thiophenes and furans. In contrast, oxidation by alkaline permanganate produces polycarboxylic and cycloalkanoic acids. The oxidant attacks heterocyclic systems primarily in the 1,1' position, so that the heteroatom is lost. However, oxidation of alicyclic systems must not necessarily involve degradation of the ring: cycloalkanoic acids were identified in appreciable amounts in the oxidation products of some kerogens (*e.g.*, Messel shale kerogen).³⁶ Nevertheless, oxidative degradation allows the genesis of cyclic and heterocyclic structures to be checked. Most probably they were generated by the Tissot–Welte mechanism which involved diagenetic cyclization and heteroincorporation (S and O) of terminally bifunctionalized aliphatic chains.⁴² Assuming that this interpretation is correct, the following postulates may be proposed:

(a) similar ranges and distributions of identified alkane-tri- and tetracarboxylic acids suggest that these acids are oxidation products of cycloalkane and/or heterocyclic structures which originated from the same source and were generated in the same way; and

(b) a similarity in the distributions of these acids and the α,ω -alkane-dicarboxylic acids (with or without a shift in the number of carbon atoms) suggests the dicarboxylic acids precursors to have been simultaneously the substrate in the diagenetic cyclization or S- and O-incorporation.

The alkane-tri- and tetracarboxylic acids identified in the oxidation products of Pumpherston shale kerogen were neither comparable with, nor resembled the identified α,ω -alkane-dicarboxylic acids, in contrast to the corresponding acids obtained by degradation or Aleksinac,³⁶ kukersite,^{23,36} and particularly Messel shale kerogens.³⁶ It is therefore suggested that at least one part of the alicyclic structures in Pumpherston shale kerogen represents inherited components of precursor algal tissues, since in addition to algaenans, which are preserved almost unchanged, lipidic constituents of algal tissues may also participate in the formation of kerogens. The functional groups containing lipids synthesized by *B. braunii* B alga (Table I), contain cyclic and possibly bicyclic botryococcenes as well,¹⁶ which is exclusive to *B. braunii* B. Regardless of the manner of incorporation of the latter cyclic hydrocarbons in the polymeric matrix, this process surely does not involve ring cleavage. In the oxidation products, such structural elements appear as polycarboxylic acids. The presence of this type of preserved alicyclic structural elements in Pumpherston shale kerogen does not necessarily exclude the possibility that some of the alicyclic and/or heterocyclic structures were generated by the Tissot–Welte mechanism. However, in such a case, the observed conformities in the distributions of the alkane-tri- and tetracarboxylic acids, on the one hand, and the polycarboxylic and dicarboxylic acids, on the other, would not be expected.

Branched acids

Isoprenoid acids are not always observed in the oxidation products of kerogens. If found, their yields are generally very low. The source of these acids are

isoprenoid structures. It is believed that kerogen isoprenoid structural elements mainly originate from carotenoids and chlorophyll, the tissues of aquatic photosynthetic organisms, as well as tocopherols from higher continental plants, which are bound to the matrix by C–C or C–O bonds according to the Tissot–Welte mechanism.⁴³ However, botryococcenes, the soluble lipids of *B. braunii* B alga, are also composed of unsaturated isoprenoid hydrocarbons. Moreover, they involve a variety of carotenoids (Table I). Due to the fact that botryococcenes are one of the direct precursors in the biosynthesis of algaenan,⁴⁴ it may be supposed that their isoprenoid character might have been conserved in the resistant biopolymer, probably to a minimal degree. Consequently, the isoprenoid acid identified in the oxidation products of Pumpherston shale kerogen may be an indicator of the role of continental flora in its genesis, but also of the presence of *B. braunii* B alga remnants, *i.e.*, of its fossilized algaenans.

The isolated *antieso*- and other branched acids, the yields of which were low, could be related to bacterial biomass providing they originated from preserved biostructures. However, these acids may just qualitatively represent the contribution of bacterial tissues in the formation of Pumpherston shale kerogen.

CONCLUSIONS

The optimised stepwise alkaline permanganate degradation was shown to have potentials for elucidating the genesis pathway and the origin of kerogens, and, specifically for revealing preserved precursors in a kerogen matrix.

Pumpherston shale kerogen, the substrate in this study, was found to be of mixed origin, *i.e.*, to contain both inherited resistant algal structures, as well as resistant biomacromolecular components of continental flora (cutans). Hence, it is suggested that this kerogen or parts of it were formed by the “selective preservation” pathway. The following evidence was observed.

The similarities between the pyrograms of the low polar pyrolysates of *B. braunii* A and B algaenans, and the range, composition and distribution of α,ω -alkane-dicarboxylic acids obtained by oxidative degradation of Pumpherston shale kerogen, indicate that both the mentioned *B. braunii* races could be the precursor algal population of this kerogen. A direct proof for the involvement of *B. braunii* B algaenan could be the C₁₁ maximum in the α,ω -alkane-dicarboxylic acids series, the short alkyl substituents in the kerogen matrix manifested by the high degradation losses, and the discordance between the alkane-polycarboxylic and the α,ω -alkane-dicarboxylic acids in the oxidation products. The large amount of degradation residue, resistant towards permanganate, which most probably contained aromatic etheral bonds originating from phenoxy etheral lipids characteristic of *B. braunii* A alga, may be taken as an indicator of *B. braunii* A algal precursors.

The aliphatic skeleton of the macromolecular network of Pumpherston shale kerogen is suggested to be mostly polymethylenic, less polyalkyl-etheral or

α,ω -disubstituted. Reliable evaluation of the alkyl substitution was not expected, especially up to C₂₀₍₂₂₎ range, since the oxidation products of hydrolyzed esters inherited from the biosphere were supposed to be competitive.

Branched acids, including iso- and *anteiso*- acids, are presumably related to bacterial biomass as a precursor of Pumpherson shale kerogen. On the other hand, the identified isoprenoid acid may originate from both aquatic organisms and terrestrial plants, but also from botryococenes and carotenoids characteristic of *B. braunii* B algal lipids.

Based on the low yield of aromatic acids in the oxidation products, the share of aromatic structures in Pumpherson shale kerogen seems to be small. However, such an assumption is not necessarily correct bearing in mind the high degradation residue and its possible nature.

The optimised stepwise alkaline permanganate degradation of kerogens may lead to reliable quantification of aliphatic constituents (alkyl substituents + esters, polymethylenic and α,ω -disubstituted aliphatic chains), alicyclic and heterocyclic structures, and aromatic moieties in the kerogen matrix, provided the yield of identifiable products is high and the degradation losses and the resistant residue are small. As this was not the case in the degradation of Pumpherson shale kerogen, quantitative evaluations were only partial.

ИЗВОД

ОЧУВАНЕ ПРЕКУРСОРСКЕ СТРУКТУРЕ У КЕРОГЕНУ ШКРИЉЦА PUMPHERSTON ДОКАЗАНЕ ОКСИДАТИВНОМ ДЕГРАДАЦИЈОМ

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Доказано је да се оптимизованом ступњевитом деградацијом керогена алкалним перманганатом могу открити очуване специфичне прекурсорске структуре у макромолекуларним, хетерогеним матрицама керогена, чиме се доприноси осветљавању путева генезе керогена и дефинисању његовог порекла. У овом раду испитиван је кероген шкриљца Pumpherson. Добијени су докази да је овај кероген мешовитог порекла. Нађено је да садржи наслеђене постојане специфичне биомакромолекуларне алгалне структуре (алгаенане) алге *B. braunii* А и Б, као и отпорне биомакромолекуларне састојке кутикула континенталне флоре (кутане). На основу резултата истраживања може се претпоставити да је кероген памферстонског шкриљца, или бар неки његови делови, постао механизмом селективног очувања.

(Примљено 6. децембра 2003)

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