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Computational chemical investigation into isorenieratene cyclisation

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Abstract

In sediments, the diaromatic carotenoid isorenieratene can undergo a wide range of molecular transformations, including cyclisation reactions via a Diels–Alder mechanism resulting in either mono- and diaromatic compounds or tetracyclic isorenieratene derivatives. To study these complex diagenetic pathways, we have used molecular dynamics simulations, in concert with a newly developed reactive hydrocarbon force field (ReaxFF). This allows us to simulate the entire cyclisation pathway, including both stable intermediates as well as energy barriers related to transition states. Our simulations indicate that the formation of tetracyclic isorenieratene derivatives is likely to occur via an A-ring initiated reaction mechanism, as the reaction product resulting from A-ring closure is more stable than that derived from B-ring closure. Furthermore, the A-ring initiated tetracyclisation pathways contain one fewer high-energy hydrogen shift step than their B-ring initiated counterparts, indicating that B-ring initiated cyclisation is more likely to result in the formation of monoaromatic compounds. These observations are in excellent agreement with observed distributions of isorenieratene derivatives in sediments and provide a better understanding of the complex diagenetic pathways of isorenieratene.

1. Introduction

Isorenieratene (**I**) is a carotenoid uniquely biosynthesised by the brown-coloured strain of the green photosynthetic sulphur bacteria (Chlorobiaceae). The habitat requirements of these bacteria—strictly anoxic, sulphide-containing water at extremely low light intensities—makes isorenieratene a useful biomarker for euxinic water columns extending into the photic zone (e.g. Sinninghe Damsté et al., 1993; Repeta, 1993).

Upon incorporation in sediments, isorenieratene undergoes a wide range of molecular transformations. The molecular transformations not involving molecular fragmentation or sulphurisation can be classified into three groups, (1) hydrogenation of double bonds, which could eventually lead to isorenieratane (**II**) (Schaeflé, 1977; Hartgers et al., 1994), (2) cyclisation by means of Diels–Alder reactions, resulting in the formation of mono-, di-, tri- and finally tetracyclic[2] isorenieratene derivatives observed in sediments (Grice et al., 1996;

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[2] In fact tri-, tetra, penta and hexacyclic, but we ignore the C_1-C_6 and $C_{1'}$-$C_{6'}$ aromatic rings in our nomenclature.

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Koopmans et al., 1996; Sinninghe Damsté et al., 2001) and (3) hydrogen shifts resulting in double-bond isomerisation, which lead, in concert with the cyclisation and hydrogenation reactions, to the formation of aromatised sedimentary isorenieratene derivatives (e.g. **III–VI**, Sinninghe Damsté et al., 1995, 2001; Koopmans et al., 1996). Molecular transformations involving fragmentation include expulsion of *m*-xylene and toluene (Byers and Erdman, 1981; Jiang and Fowler, 1986; Schoell et al., 1994; Koopmans et al., 1996, 1997; Sinninghe Damsté et al., 1997) and cleavage of C-C single bonds as evidenced by the presence of a wide variety of short-chain isorenieratene derivatives in sediments, including aryl isoprenoids (Summons and Powell, 1987; Requejo et al., 1992; Hartgers et al., 1994).

Focusing on the molecular transformations not involving C–C-bond cleavage or sulphurisation, the wide range of isorenieratene derivatives observed in sediments suggests that the three types of molecular transformation (hydrogenation, cyclisation and aromatisation) are in competition with each other. Each of these transformation types has a distinct chemical nature. The hydrogenation reaction requires the presence of a hydrogen donor, making it strictly intermolecular. The hydrogen shift reaction is in principle intramolecular, although its reaction rates could be affected by the presence of co-reactants stabilising reaction intermediates. Finally, the Diels–Alder cyclisation reaction is strictly intramolecular and, due to the concerted nature of this reaction, is expected to have a much lower energy barrier than either the hydrogenation or hydrogen shift reaction. These differences in chemical nature suggest that environmental parameters, such as temperature and concentration of co-reactants, will have a strong effect on the relative prevalence of these three transformation types. Analyses of sedimentary samples with different thermal histories support these suggestions, as a sample from a thermally immature Mediterranean sapropel shows significant amounts of tetracyclic compounds, while a Kimmeridge Clay sample subjected to higher temperatures upon burial shows predominantly mono- and bicyclic isorenieratene derivatives (Sinninghe Damsté et al., 2001).

Understanding the effect of the depositional environment on the distributions of isorenieratene derivatives requires an analysis of the diagenetic pathways leading to these derivatives. This analysis is complicated by the fact that the derivatives observed in sediments all seem to be end-products of the diagenetic pathways, like fully hydrogenated or aromatised compounds, leaving few clues regarding the sequence of hydrogenation/ring closure/hydrogen shift reactions responsible for their formation. To help overcome these complications we have performed a series of computational chemical simulations. In earlier work (Sinninghe Damsté et al., 2001) we reported on the relative stabilities of isorenieratene and its various aromatised and non-aromatised tetracyclic derivatives, identifying the thermodynamic driving force for cyclisation and subsequent aromatisation reactions. Furthermore, by calculating the stabilities of various intermediates in the first Diels–Alder cyclisation step (single, double and triple-*cis* configurations of isorenieratene) we obtained clues regarding the sequence of the first steps of the isorenieratene diagenetic pathway. In this earlier work, the MM3-force field for conjugated hydrocarbons was used (Allinger et al., 1989). This method allows the calculation of the thermodynamic stability of isorenieratene and its derivatives but cannot be used to calculate the energy barriers for the various reaction steps, as it can only be applied to stable hydrocarbons (i.e. hydrocarbons without dissociating bonds). These energy barriers can have a profound impact on the sedimentary distribution of isorenieratene derivatives, as high energy barriers could render reaction pathways leading to thermodynamically stable compounds inaccessible. By using the MM3-method, these important reaction kinetics issues could not be addressed in our earlier study. However, the choice for using the thus restricted MM3-method was based on practical considerations, as quantumchemical computational chemical methods that would allow thermodynamic analysis of the reaction intermediates associated with these energy barriers require unrealistically long computational times for compounds of this size. Semi-empirical computational methods allow analysis of a specific reaction step associated with carotenoid chemistry (Kuki et al., 1991; Doering et al., 1995) but are still too time-consuming for an analysis of the full isorenieratene cyclisation pathway. Recently, these computational chemical limitations have been resolved by the development of a reactive force field for hydrocarbons (ReaxFF; van Duin et al., 2001). This empirical method allows simulation of many types of chemical reactions, including bond dissociation and concerted Diels–Alder reactions, using computer times that are magnitudes lower than quantumchemical or even semi-empirical methods. This allows us to expand on our earlier MM3-work by determining the energy barriers for the reaction steps leading towards isorenieratene cyclisation, addressing potential kinetic controls imposed on the diagenetic pathway by unstable reaction intermediates.

The full cyclisation pathway of isorenieratene involves only two of the three molecular transformation types discussed earlier. Diels–Alder reactions are responsible for the cyclisation. As these pericyclic reactions are of the $4+2$ type, which are, in contrast to the $2+2$ cycloadditions, allowed in the electronic ground state, these reactions should be thermal, not requiring light to access excited electronic states. After such a Diels–Alder cyclisation step, the remaining double bonds may not be properly configured for subsequent

cyclisations, thus requiring hydrogen shifts to reposition them for the next concerted reaction step. As such, full cyclisation is an interplay between Diels–Alder and hydrogen shift reactions. The third molecular transformation type, double bond hydrogenation, although not part of the full cyclisation pathway and as such not investigated here, composes an important restraint on the full cyclisation pathway as any double bond hydrogenation immediately disqualifies full cyclisation as a diagenetic option, leaving only hydrogenated or mono- or bicyclic aromatic compounds as potential diagenetic endpoints. Other diagenetic modifications, like C–C cleavage or incorporation of sulphur, are likely to have a similar effect as double bond hydrogenation in that they remove full cyclisation as an option. Our approach towards studying the cyclisation of isorenieratene comprises first a computational simulation of all potential Diels–Alder cyclisation steps starting from the isorenieratene parent molecule. This should resolve the question which cyclisation step initiates the first step of the isorenieratene diagenetic pathway, allowing us to justify our computational approach by a comparison with isorenieratene derivatives observed in sediments. Thereafter, we compare the thermodynamic consequences of several diagenetic schemes, consisting of various hydrogen shift and Diels–Alder steps, eventually leading to the formation of the tetracyclic compounds. These two parts are preceded by the results of the recalculation of the MM3-data from our earlier work using the ReaxFF-force field, demonstrating that these two methods produce equivalent results for the stable isorenieratene derivatives.

In summary, we describe the application of thermodynamic data derived from computational chemical simulations, cross-checked with sedimentary compound distributions wherever possible, to illuminate the complex diagenetic fate of isorenieratene. This should increase the already substantial application of this compound and its derivatives as palaeoenvironmental indicators and also serves to demonstrate the potential of contemporary computational chemical methods for organic chemical and organic geochemical research.

2. Methods

To calculate the thermodynamic stability of the isorenieratene derivatives and to analyse the energy barriers associated with the reaction steps along the isorenieratene diagenetic pathway the ReaxFF reactive hydrocarbon force field was used, as described by van Duin et al. (2001). Fundamentally, ReaxFF is similar to non-reactive force fields like MM3 (Allinger et al., 1989) in that the system energy is divided into partial energy contributions associated with, amongst others, bond stretching, valence angle bending and non-bonded van der Waals and Coulomb interactions. A new feature of the ReaxFF force field is that by using a bond order approach and by rigorous implementation of these bond orders in all partial energy contributions a smooth description of bond formation and dissociation is obtained, which has been scrutinised by comparison with quantumchemical data. A molecular mechanics scheme (e.g. Burkert and Allinger, 1982) was used to determine the minimum energy configuration for the isorenieratene derivatives. By adding a heat increment proportional to the number of carbon and hydrogen atoms, corrections for translational and rotational modes and corrections for pV-work this minimum system energy was translated into heat of formation data (van Duin et al., 2001). ReaxFF was parameterised, amongst others, against a large set of experimentally derived heats of formation for both conjugated and non-conjugated hydrocarbons. The average deviation between ReaxFF and experiment, 2.8 kcal/mol for non-conjugated and 2.9 kcal/mol for conjugated systems (van Duin et al., 2001), indicates the level of accuracy we can obtain with this method. To determine the reaction energy barriers, a molecular dynamics (MD; e.g. Allen and Tildesley, 1987; Frenkel and Smit, 1996) method was used in association with a restraint method. Firstly, the unrestrained molecule was equilibrated at a temperature of 298 K, using a velocity scaling method as described by Berendsen et al. (1984) to control the temperature. Subsequently, a set of sliding restraints [Eq. (1)] were added to the ReaxFF-system energy description, forcing the molecule to transform its configuration alongside the reaction coordinate. The isorenieratene reactions were forced by gradually shifting the restraint distance $R_{\text{restraint}}$ from the interatomic distance r_{ij} in the reactant to that in the reaction product (Fig. 1).

$$E_{\text{restraint}} = k_1 \cdot \left\{ 1 - \exp\left(-k_2 \cdot \left(r_{ij} - R_{\text{restraint}} \right)^2 \right) \right\} \qquad (1)$$

By performing this gradual restrained transformation in concert with an MD method at $T = 298$ K, the non-reacting part of the molecule was allowed to change its conformation as a response to the reactive molecular alterations. The thermodynamic effects of the reaction, including overall reaction energy as well as energy barriers, can be determined by monitoring the system energy during the restrained MD-simulation. For the Diels–Alder reactions one restraint, between the ring-closing atoms, sufficed to force the reaction, with restraint force constants k_1 and k_2 having values of 1500 kcal/mol and 0.10 Å^{-2}, respectively. Each Diels–Alder simulation required 90,000 iterative MD steps to finish the entire cyclisation step, with each iteration simulating a time interval of 0.1 femtoseconds. This simulation requires about 90 min on a Compaq XP10000 workstation. For the hydrogen shift reactions two sliding

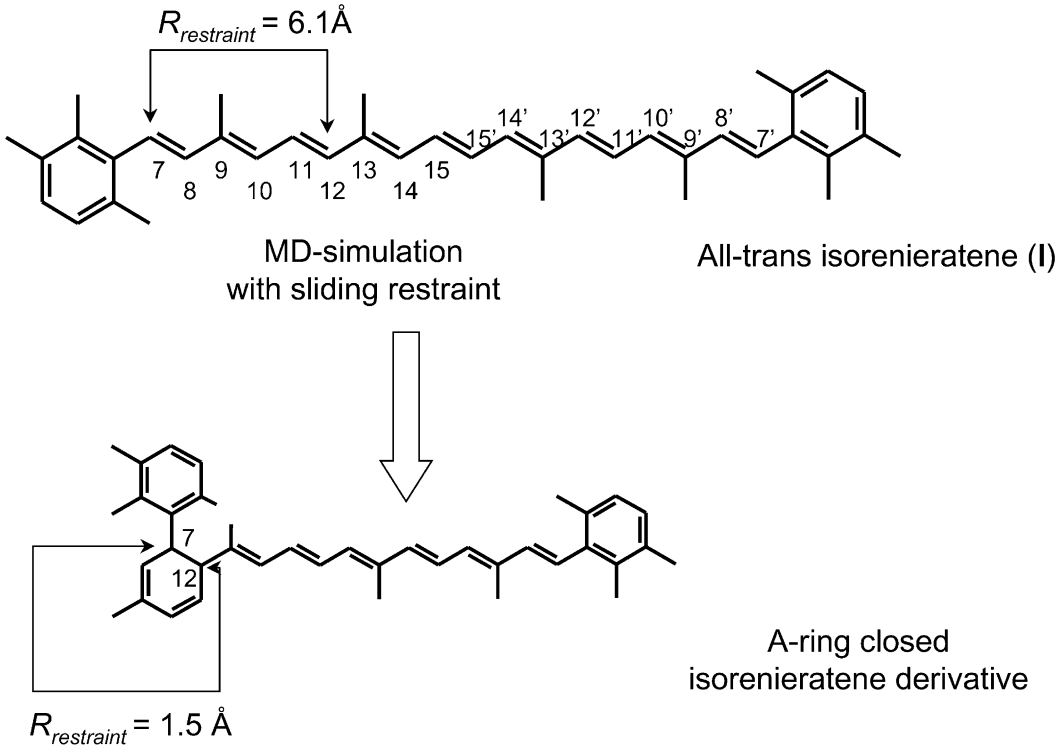

Fig. 1. Sliding restraint method for forcing the ring closure reactions in isorenieratene.

restraints were used, one between the donating carbon and the shifting hydrogen and the other between the accepting carbon and the shifting hydrogen, each simulation requiring 10,000 iterative MD-steps at 298 K with restraint force constants $k_1 = 1500$ kcal/mol and $k_2 = 0.03$ Å^{-2}. Changes in the double or single bonded nature of carbon-carbon linkages during either the Diels–Alder or hydrogen shift reactions were automatically taken into account by the ReaxFF-method. By taking block averages over intervals of 750 iterations the thermal noise was filtered out of the reaction energy profile, allowing the determination of the energy barriers for the isorenieratene cyclisation steps.

The simulations presented here were performed on a single isorenieratene molecule at zero pressure. As such, in interpreting these results one should take into account that the reaction mechanisms we find may not completely describe sedimentary events, where temperature effects, elevated pressure and the presence of co-reactants will affect isorenieratene chemistry. However, a firm understanding of the isorenieratene gas phase chemistry, to which this study aims to contribute, will greatly aid in the recognition of these temperature, pressure and composition effects on isorenieratene diagenesis.

3. Results and discussion

3.1. Comparison with MM3

In this study we used the ReaxFF-force field, because this force field can be used to simulate bond formation/dissociation processes in hydrocarbons, in contrast to the MM3 force field employed in our earlier work (Sinninghe Damsté et al., 2001). Fig. 2 compares the MM3 heat of formation data presented in our earlier study with ReaxFF heats of formation. This demonstrates that the thermodynamic driving force for isorenieratene cyclisation and subsequent aromatisation, as identified in our earlier work using the MM3-data, is also recognised by ReaxFF. In general, there is good agreement between the MM3 and ReaxFF data, the one significant difference being that ReaxFF predicts a systematically lower stability for the conjugated tetracyclics, but as a group these compounds are still more stable than the non-conjugated tetracyclics.

3.2. First cyclisation step

Fig. 3 shows the energy profile obtained from an MD-simulation in which a sliding restraint was applied to enforce a ring closure between C_7 and C_{12}, resulting

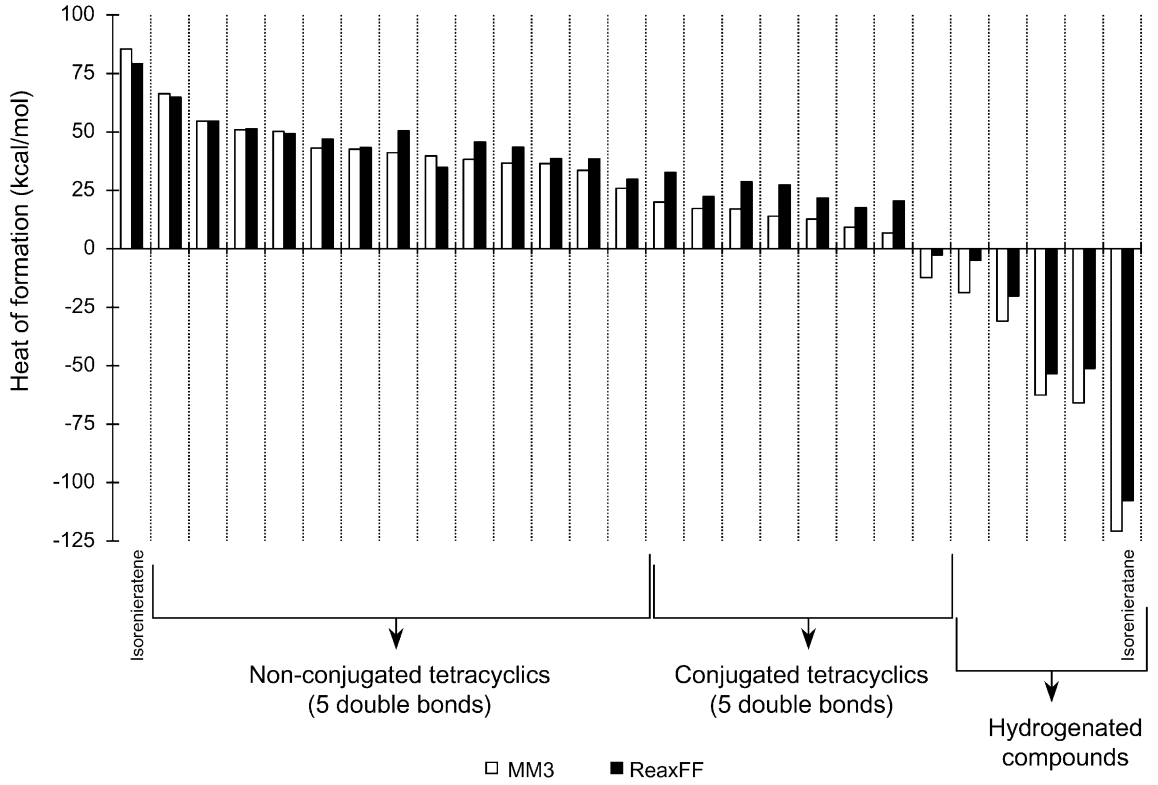

Fig. 2. Calculated heat of formation for isorenieratene and its derivatives using the MM3 and ReaxFF-force fields.

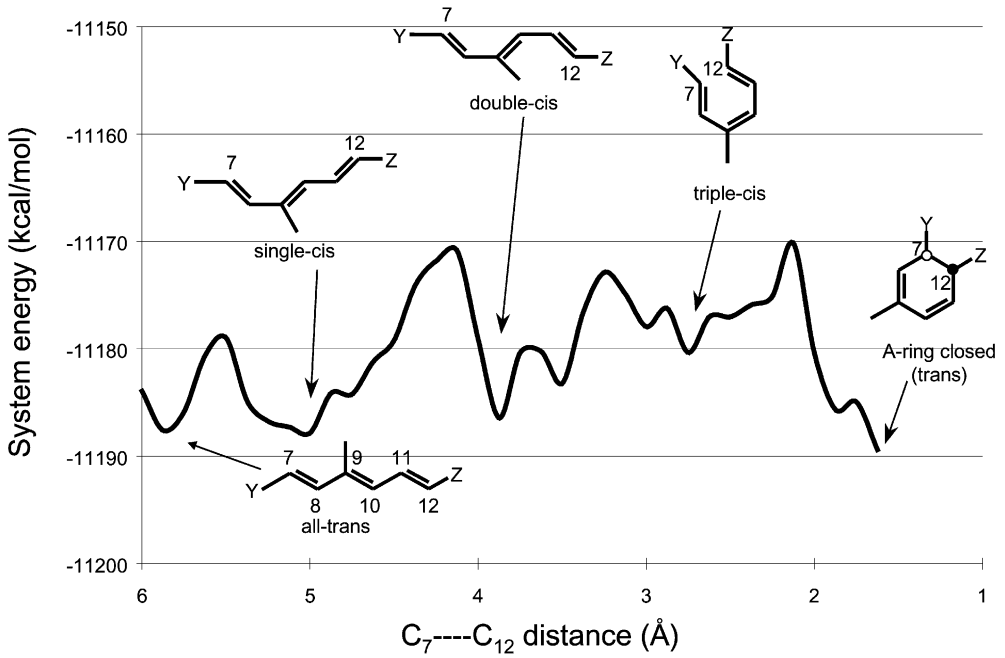

Fig. 3. Energy profile obtained from the MD-simulations on A-ring (C_7–C_{12}) closure in isorenieratene. Y depicts a trimethylphenyl-fragment, Z depicts the other non-reactive part of the isorenieratene molecule.

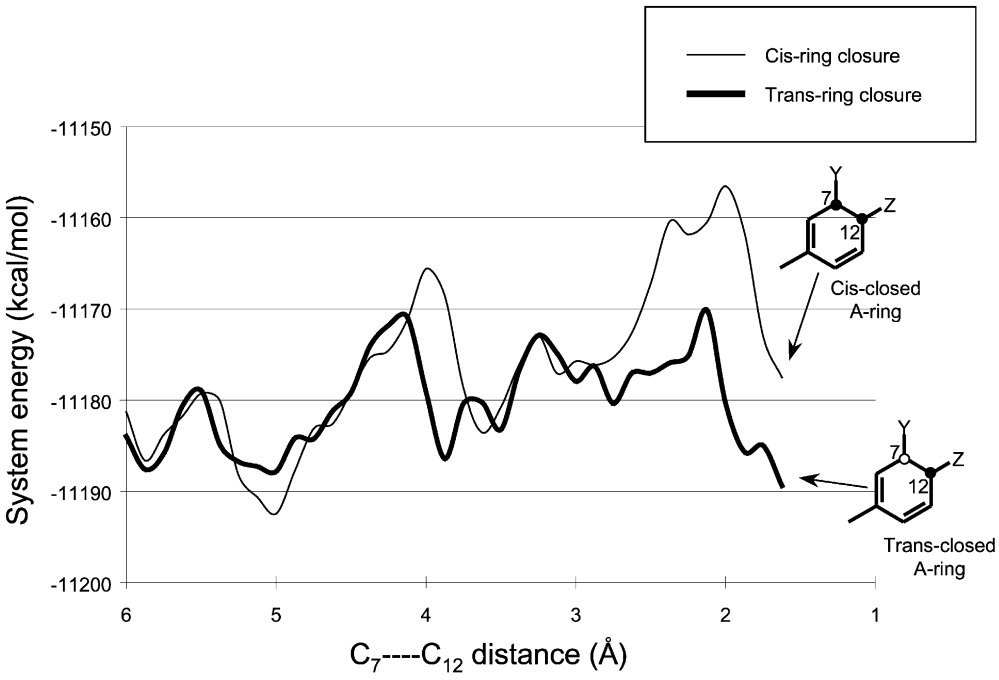

Fig. 4. Energy profiles for *cis*- and *trans*-A-ring closure in isorenieratene. Y depicts a trimethylphenyl-fragment, Z depicts the other non-reactive part of the isorenieratene molecule.

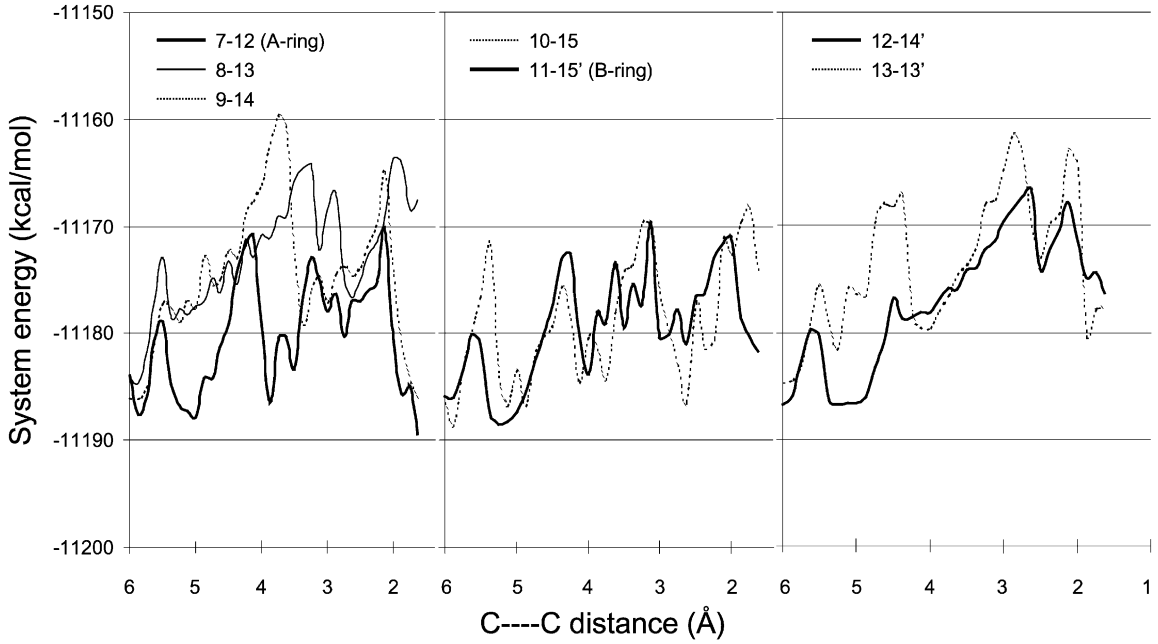

Fig. 5. Energy profiles for the seven possible first cyclisation steps in isorenieratene. For atom numbering and A/B ring definition, see Fig. 1 and the Appendix.

in the formation of an A-ring closed isorenieratene derivative (see the Appendix, compound **VII**, for a definition of A, B, C and D-rings). Five stages can be identified on the reaction coordinate, commencing with the all-*trans* conformation. With decreasing C_7—C_{12} distance the all-*trans* conformation gets transformed into, subsequently, single-, double- and triple-*cis* conformations and eventually the triple-*cis* conformation undergoes cyclisation via a Diels–Alder reaction. Each of these five stages is clearly separated from its adjoining stages by an energy barrier. The energy barrier for the Diels–Alder cyclisation step is about 10 kcal/mol, which is comparable to those observed for the *trans*/*cis* conversions. These low barriers indicate that the ring-closure reaction should proceed very fast. In addition, due to the fact that the A-ring closed compound is only marginally more stable than the all-*trans* isorenieratene parent molecule, each of these reaction steps should be readily reversible, as even the largest energy barrier on the reversed pathway, associated with the retro Diels–Alder reaction of the A-ring closed compound to the triple-*cis* conformation, amounts to only 20 kcal/mol.

Fig. 4 compares the energy profile of the *trans*-A ring closure, as depicted earlier in Fig. 3, with that of a *cis*-ring closure. This demonstrates that the energy barrier for the final stage of the reaction, the Diels–Alder cyclisation step, is significantly higher for the *cis*-ring closure and also leads to a less stable end product. This indicates that *trans*-ring closure should be the prevalent cyclisation pathway and, for that reason, we will only consider *trans*-cyclisations from now on.

Due to the 2-fold symmetry in isorenieratene, only seven options for the first cyclisation step exist. Comparison between the energy profiles of these seven cyclisation pathways (Fig. 5) shows that some of these ring closure reactions are unlikely to occur, either due to high energy barriers (9-14 and 13-13′ closure) or because they result in unstable end products (8-13 closure). Judging from the energy profiles in Fig. 5, 7-12 and 11-15′ring closure should be the major cyclisations products, with 10-15 and 12-14′ ring closures forming minor products. This fits very well with suits of monoaromatic isorenieratene derivatives observed in sediments (Sinninghe Damsté et al., 2001): A-ring and

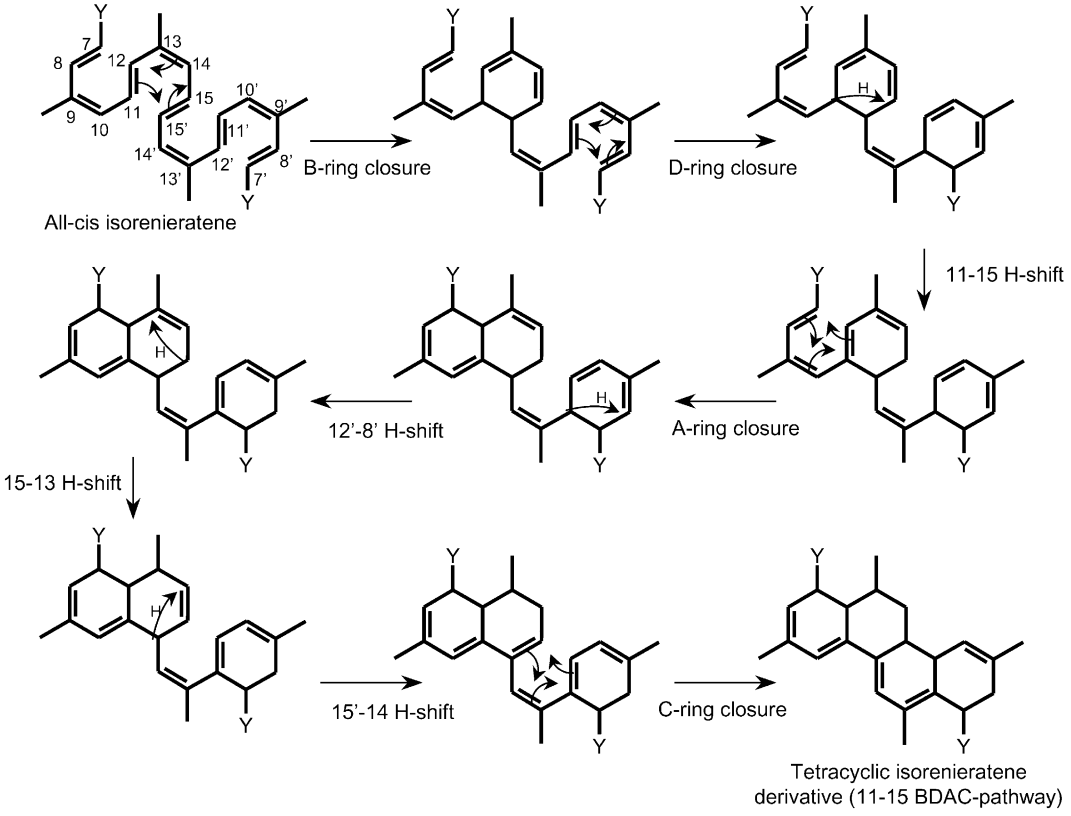

Fig. 6. Diagenetic pathway for the formation of tetracyclic isorenieratene derivatives using subsequent B, D, A and C-ring closure steps. If required, these Diels–Alder ring closure steps are followed by hydrogen shift reactions to get the double bonds in the appropriate positions for the next ring closure step. Y depicts a trimethylphenyl-fragment (see Appendix). An all-*cis* configuration is used only for drawing convenience; the all-*trans* configuration is thermodynamically more stable and, as such, is a more likely configuration for the parent isorenieratene molecule.

B-ring aromatics (**III** and **IV**), associated with 7-12 and 11-15′-ring closure, are prevalent, while lower concentrations of compounds **V** and **VI**, associated with 10-15 and 12-14′ ring closures, are observed. Furthermore, no monoaromatic isorenieratene derivatives related to 8-13, 9-14 or 13-13′ ring closure have been observed in sediments.

One major discrepancy between the distribution of sedimentary monoaromatic isorenieratene derivatives and the simulation results is that the product of a 7-12 (A)-ring closure is thermodynamically more stable than that of a 11-15′ (B)-ring closure (Fig. 5), whereas sedimentary data show a 60/40 ratio of B-ring over A-ring aromatics (Sinninghe Damsté et al., 2001). The validity of the ReaxFF-predicted A-ring preference was examined by re-determination of the stabilities of the 7-12 and 11-15′ ring closure products using MM3 (Allinger et al., 1989) and MOPAC/PM3 (Dewar and Thiel, 1977; Stewart, 1989). Both these computational methods confirmed the ReaxFF stability sequence. Assuming, for that reason, that the ReaxFF-predictions are valid, this indicates that B/A ring dominance in the monoaromatic fraction is not only controlled by the rate of the first cyclisation steps but also by later diagenetic reactions. One of these reactions could be preferential degradation of A-ring aromatics over B-ring aromatics. However, both ReaxFF and MM3 predict compound **III** to be thermodynamically significantly more stable than **IV** ($\Delta H_f^{III}(MM3) = -69.6$ kcal/mol; $\Delta H_f^{IV}(MM3) = -65.9$ kcal/mol), making degradation an improbable explanation for B-ring aromatic predominance. This stability difference of about 4 kcal/mol in favour of **III** makes it also very improbable that entropy differences between **III** and **IV** could, at sediment temperatures, shift the balance towards the B-ring aromatics.

3.3. Full cyclisation pathways

After the first cyclisation step, a sequence of Diels–Alder reactions and hydrogen shifts is required to transform monocyclic isorenieratene into its tetracyclic derivatives. The hydrogen shift steps reconfigure the double bond positions so that subsequent Diels–Alder reactions can occur. Fig. 6 shows a possible full cyclisation pathway, involving, in this order, B, D, A and C ring closures. The pathway depicted in Fig. 6 involves four Diels–Alder and four hydrogen shift steps. By replacing the 11-15 hydrogen shift step with a 11-13 hydrogen shift the same tetracyclic compound can be obtained requiring one less hydrogen shift. Fig. 7 compares the energy profiles of these two possible BDAC cyclisation pathways. A number of conclusions can be drawn from these energy profiles. First, they demonstrate that the energy barriers for the hydrogen-shift reactions are at least 20 kcal/mol higher than those for the Diels Alder cyclisation or *trans*/*cis* transformation steps. This indicates

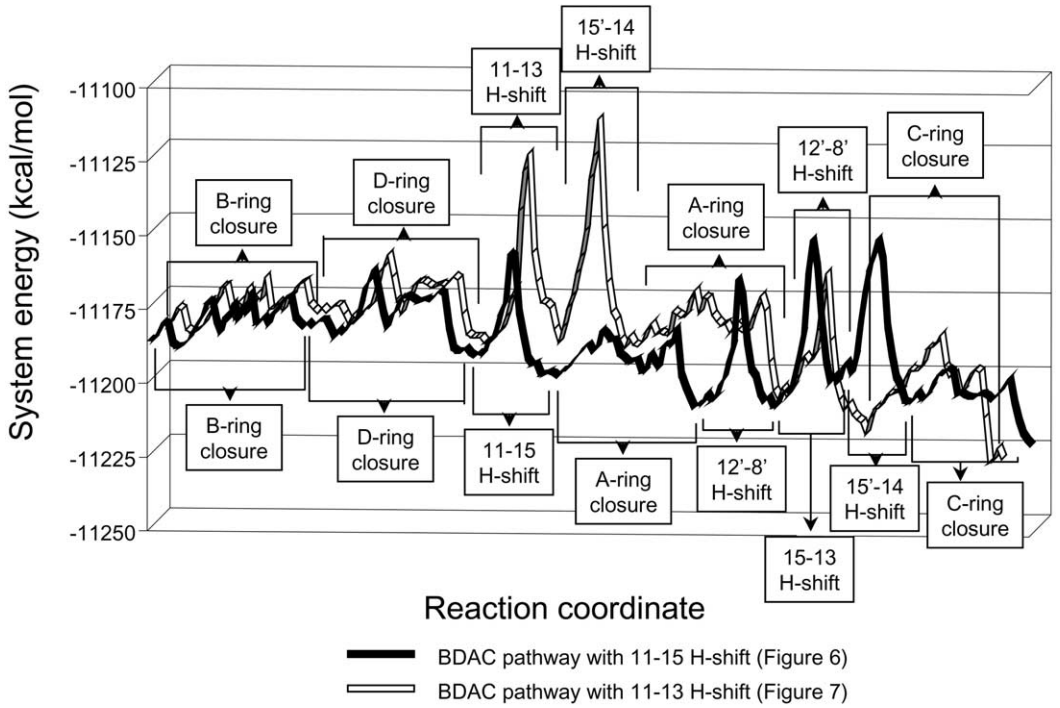

Fig. 7. Comparison between the energy profiles for the 11-15 hydrogen shift BDAC-ring closure pathway (Fig. 6) and the 11-13 hydrogen shift BDAC-ring closure pathway.

that the hydrogen-shift reactions are the rate-determining steps in the formation of the tetracyclic isorenieratene derivatives. Because of this, one would expect that the 11-13 BDAC-pathway is preferred over the 11-15 BDAC-pathway (Fig. 6), because the latter pathway involves one additional hydrogen-shift step. However, as Fig. 7 shows, the 11-13 and subsequent 15′-14 hydrogen-shifts have energy barriers about 25 kcal/mol higher than any of the hydrogen-shifts encountered on the 11-15 BDAC pathway. This means that at sedimentary conditions (long reaction times and relatively low temperatures) the 11-15 BDAC pathway will be preferred over the 11-13 BDAC pathway, despite the additional hydrogen-shift step.

A comparison of a full cyclisation pathway commencing with A-ring closure, like the ABCD pathway in Fig. 8, to the B-ring initiated pathway in Fig. 6 shows that the A-ring initiated pathway requires one fewer hydrogen shift step. When discounting high-energy shortcuts like the 11-13 BDAC pathway discussed earlier, this is a recurring theme when comparing A- to B-ring initiated full cyclisation pathways (Table 1). With the exception of the ACBD and ACDB-pathways, which are equivalent to the BDAC and BDCA pathways, all A-initiated pathways require one fewer hydrogen shift step than the B-ring pathways. Fig. 9 shows that the hydrogen shift energy barriers in the A-initiated cyclisation pathways have energy barriers similar to or lower than those in the B-initiated pathways. This, combined with the conclusion from the energy profiles in Figs. 7 and 9 that hydrogen shifts are the rate-determining steps in the full cyclisation pathway, suggests that the A-ring initiated full cyclisations, containing fewer of these hydrogen shift steps, should proceed at a considerably higher rate than those commencing with B-ring cyclisation. Fig. 9 also shows that, regardless of the cyclisation pathway, tetracyclisation is thermodynamically favourable, as the tetracyclic reaction products are about 25 kcal/mol more stable than *trans*-isorenieratene. This confirms the previously identified thermodynamic driving force for isorenieratene diagenesis (Sinninghe Damsté et al., 2001; Fig. 1).

Having thus established A-ring initiated cyclisation as the dominant pathway for the formation of tetracyclic

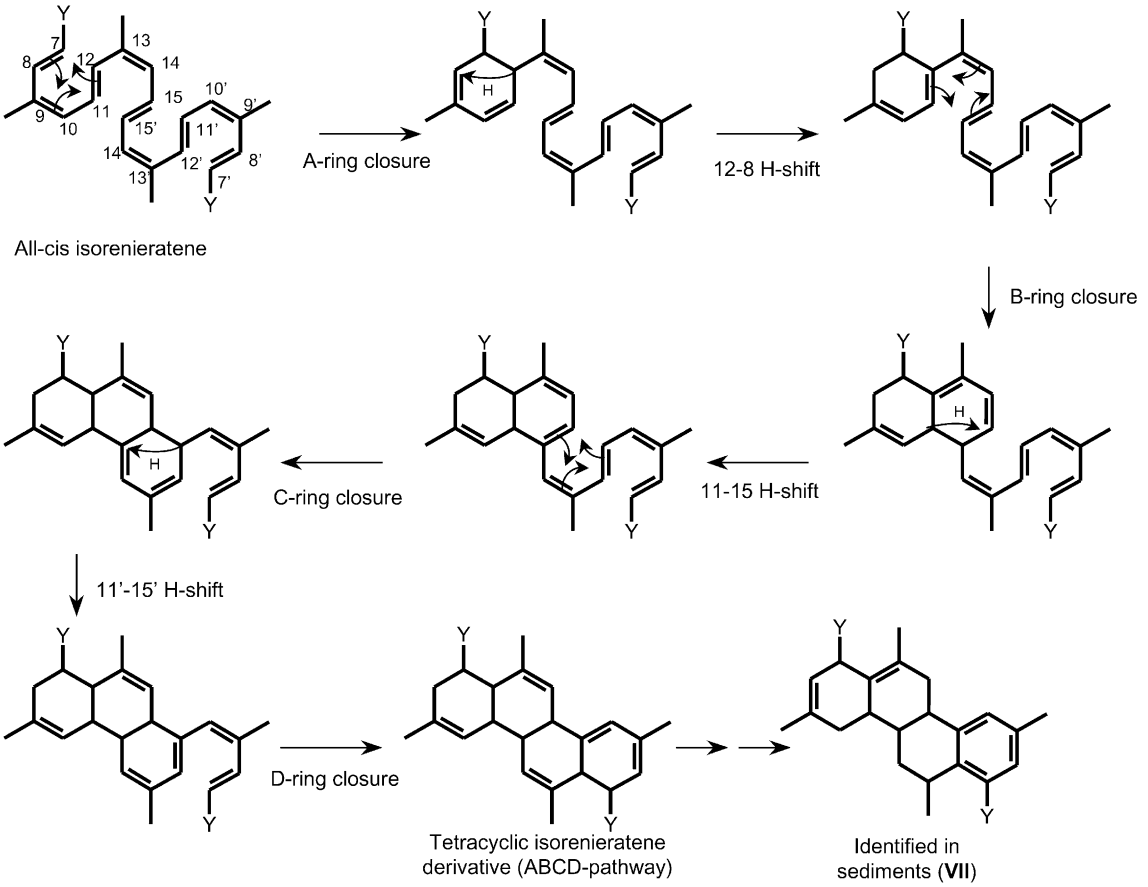

Fig. 8. Diagenetic pathway for the formation of tetracyclic isorenieratene derivatives using subsequent ABCD-ring closure steps. Y depicts a trimethylphenyl-fragment (see Appendix).

Table 1
Number of Diels–Alder and hydrogen shift steps required to complete full isorenieratene cyclisation using different pathways. Assuming exclusive *trans*-cyclisations, 12 different cyclisation pathways are possible. Due to the two-fold symmetry in isorenieratene, some of these pathways are equivalent (as noted), involving straightforward juxtaposition of reaction steps

Cyclisation sequence	Diels–Alder steps	Hydrogen shift steps
ABCD (=ABDC)	4	3
ADBC (=ADCB)	4	3
BADC (=BACD)	4	4
BCAD (=BCDA)	4	4
BDAC (=BDCA, ACBD and ACDB)	4	4

isorenieratene derivatives, the question remains whether the ABCD- (Fig. 9a) or the ADBC-pathway (Fig. 9b) is more likely to occur. The ADBC-cyclisation product is somewhat more stable than the ABCD-derived tetracyclic compound. However, the cyclisation steps and, more importantly, the hydrogen shift steps in the ADBC-pathway have energy barriers which are consistently higher by about 5 kcal/mol than those in the ABCD-pathway. Although these margins do not rule out the ADBC-pathway completely, they do suggest that the ABCD-pathway should prevail, especially in low-temperature sediments. Encouragingly, this seems in line with sedimentary evidence, as only a few straightforward, and thermodynamically favourable (Sinninghe Damsté et al., 2001), double bond rearrangements separate the product of ABCD-cyclisation from **VII**, which is the only non-hydrogenated aromatic tetracyclic isorenieratene derivative observed so far in sediments (Bosch et al., 1998; Passier et al., 1999; Sinninghe Damsté et al., 2001).

Returning to the discussion at the end of the previous section, a full cyclisation pathway dominated by A-ring initiation could also explain the apparent contradiction between sedimentary predominance of B-ring over A-ring monoaromatics and the preference of A- over B-ring closure as a result of the first cyclisation step. Full cyclisation requires that none of the double bonds in isorenieratene gets hydrogenated, a process that is in continual competition with the cyclisation reactions. Since full cyclisation is going to be slower when initiated with a B- instead of an A-ring cyclisation, hydrogenation of a double bond before reaching the tetracyclic

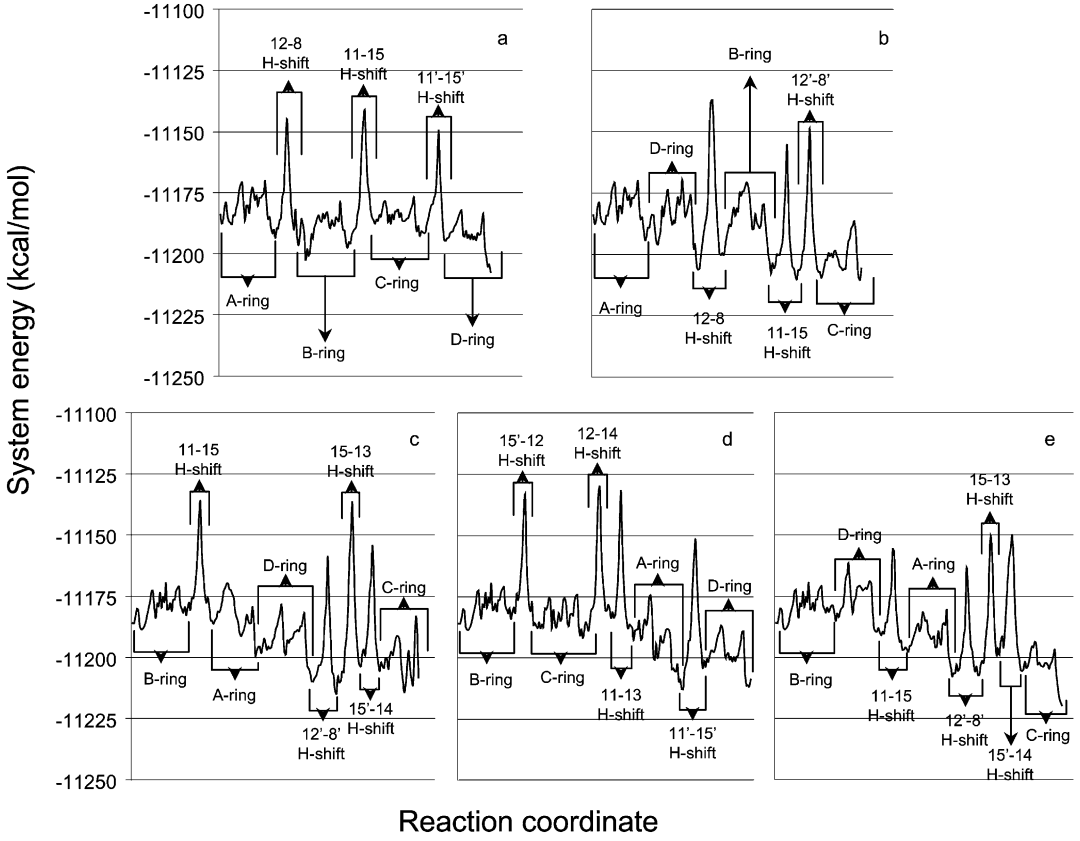

Fig. 9. Energy profiles for the ABCD- (a), ADBC- (b), BADC- (c), BCAD- (d) and BDAC-ring (e) closure pathways.

stage is more likely after an initial B-ring closure. After such a hydrogenation, monoaromatic compounds like **IV** and related diaromatic compounds are virtually the only possible diagenetic end products (excluding those products formed after C–C bond cleavage). As such, this apparent mismatch between our computational results and sedimentary isorenieratene derivative distributions can be explained by competition between full cyclisation and hydrogenation/aromatisation pathways.

As demonstrated in this work, the different reaction steps partaking in isorenieratene diagenesis have distinctly different characters, ranging from intramolecular Diels–Alder reaction with low energy barriers, which should dominate at low sediment temperatures, to hydrogen shift steps with higher energy barriers, which could potentially have a partly intermolecular character due to catalysing co-reactants, and finally hydrogenation reactions, which are by definition intermolecular and will very probably have higher energy barriers than the Diels–Alder cyclisations. The continuous competition between these reaction steps, resulting in diagenetic pathways with often clearly different end products, makes the isorenieratene derivatives distribution into a potentially powerful, albeit complicated, indicator for depositional environment and sediment temperature history. Further study, including expansion of the simulated gas-phase chemistry to condensed phase, including the effects of intramolecular reactions, is required to fully unlock the biomarker potential of isorenieratene derivatives in sediments. Ongoing developments in ReaxFF reactive force fields will also allow for future studies on isorenieratene reactions with heteroatoms (notably sulphur), which should allow us to expand on the isorenieratene diagenetic scheme sketched out here.

4. Conclusions

Molecular dynamics simulations, using the recently developed reactive hydrocarbon force field ReaxFF, have been used to study the isomeric conversion of isorenieratene (**I**) into its tetracyclic derivatives (e.g. **VII**). These simulations indicate that A-ring closure is preferred as the initiating step over B-ring closure. Furthermore, with regard to the full cyclisation pathway, comprising a combination of Diels–Alder cyclisations and hydrogen shift steps, we find that the hydrogen shift steps have considerably higher energy barriers, identifying them as the rate determining steps in this pathway. Since A-ring initiated cyclisation pathways contain only three hydrogen shift steps, in contrast to their B-ring initiated counterparts that contain four, this suggests that tetracyclisation of isorenieratene predominantly follows the A-ring initiated pathway. B-ring initiated cyclisations are more likely to result in formation of monoaromatic compounds (e.g **IV**), which explains the predominance of B-monoaromatics over A-monoaromatics (**III**) in sediments, despite the higher thermodynamic stability of the latter compound class.

Our simulations suggest that of the possible A-initiated cyclisation pathways, the ABCD-pathway (Fig. 8) is the most likely, which is in good agreement with the structure of the only sedimentary tetracyclic isorenieratene isomer identified so far (**VII**).

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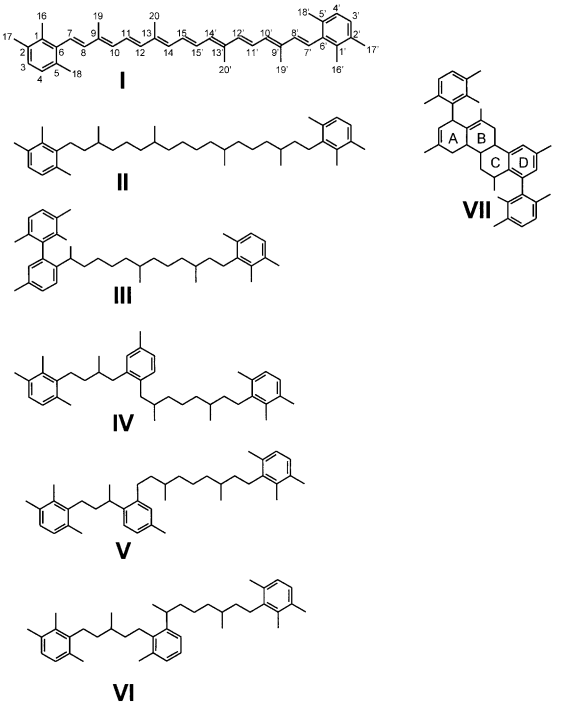
Appendix



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