Reductive Coupling of Aldehydes by H_2S in Aqueous Solutions, a C–C Bond Forming Reaction of Prebiotic Interest

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We report here a novel reductive coupling reaction of conjugated, non- or poorly enolizable aldehydes induced by H_2S and operative in aqueous solutions under prebiotically relevant conditions. This reaction leads from retinal to β-carotene, and from benzylic aldehydes to the corresponding diarylethylenes. This novel reaction also opens a new potentially prebiotic pathway leading from glyoxylic acid to various compounds that are involved in the reductive tricarboxylic acid cycle. This C–C bond forming reaction of prebiotic interest might have been operative, notably, in the sulfide-rich environments of hydrothermal vents, which have been postulated as possible sites for the first steps of organic chemical evolution.

Introduction. – Formation of C–C bonds under relevant aqueous conditions is a central question of prebiotic chemistry and is the subject of numerous studies [1]. In this context, we have discovered a novel reductive coupling reaction of conjugated aldehydes and non-enolizable aldehydes induced by H_2S and sulfides in the course of our investigations of non-biological reduction processes affecting sedimentary organic matter from anoxic settings [2]. We have investigated this reaction in detail within the frame of our study of abiotic organic synthetic processes likely to be operative under conditions prevailing in the vicinity of hydrothermal vents [3] where fluids containing high amounts of gases of prebiotic significance (*e.g.*, H_2S, CO_2, CO, H_2, ...) are emitted in the seawater [4]. This reaction, which leads from retinal to β-carotene but also from benzylic aldehydes to the corresponding diarylethylenes and from glyoxylic acid to various C_4–C_6 carboxylic acids under aqueous conditions, has a clear relevance to prebiotic chemistry. It can be considered as an 'aqueous' analog of the *McMurry* coupling reaction of conjugated carbonyl compounds which is normally performed under strictly anhydrous conditions [5].

Results and Discussion. – In typical experiments aimed at investigating the abiotic reduction reactions of prebiotic and geochemical interest induced by H_2S and sulfides on various organic compounds under aqueous conditions in the absence of transition metal catalysis, the substrate was adsorbed on *Celite* to ensure a good dispersion, and the solid phase obtained was suspended in a 0.1M solution of NaOH in H_2O saturated with H_2S. After heating at 50 or 90° for periods extending from 7 to 15 d, the organic

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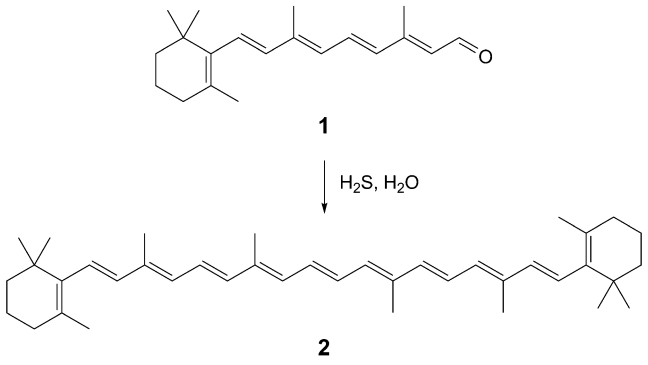
extract was analyzed. In the case of retinal (**1**), the analysis revealed the presence of a compound with an intense orange color, and which, based on the analysis by probe-MS and comparison with a reference molecule, corresponds to β-carotene (**2**; M^+ at m/z 536, fragment ion at m/z 444) (*Scheme 1*).

Scheme 1. *Formation of β-Carotene* (**2**) *by Reaction of Retinal* (**1**) *with* H_2S *under Aqueous Conditions*

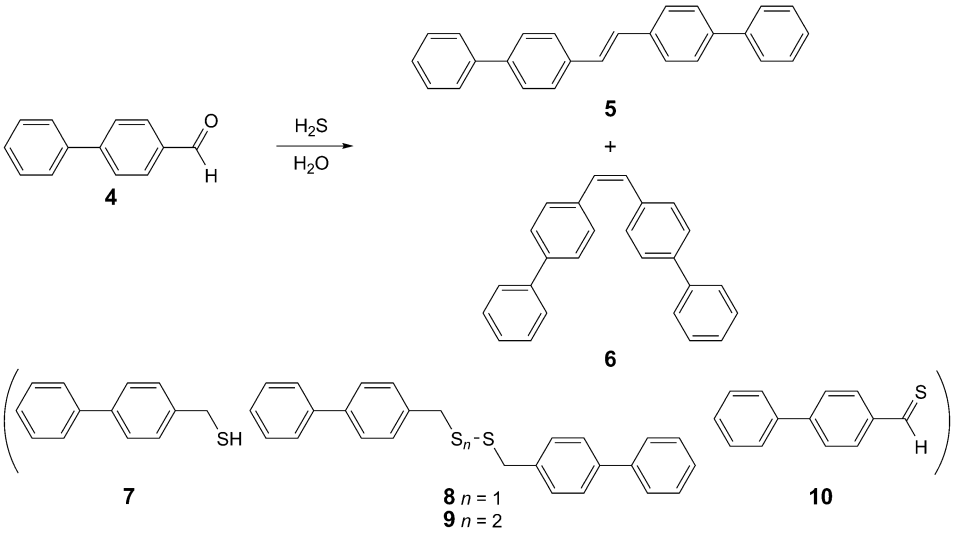


This structural hypothesis was further confirmed by reversed-phase HPLC analysis with a PDA (= photodiode array) detector which showed the occurrence of a predominant compound exhibiting the typical UV/VIS spectrum of (all-*E*)-β-carotene (**2**; λ_{max} 452 nm). A second, later eluted minor compound was also detected and was obviously an isomer of **2**. According to *Britton et al.* [6], the presence of a (*Z*)-C=C bond on a carotenoid structure induces a hypsochromic and an important hypochromic effect. The fact that the λ_{max} value (448 nm) of the minor isomer was slightly shifted as compared to that of **2** suggested that it likely corresponded to a (*Z*)-isomer of **2**. Since the C(15)–C(15′) bond has been formed during the C,C-coupling reaction, it was likely that this compound corresponded to (15(15′)*Z*)-β-carotene (**3**; see *Scheme 3*), an hypothesis also compatible with the measured λ_{max} value [7].

The ^{1}H-NMR spectrum of the major isolated orange compound was identical to that of the authentic (all-*E*)-β-carotene (**2**) [8]. Quantification of β-carotene (UV/VIS) in the various experiments showed that the yields were relatively low but increased with increasing temperature (typically in the range of 0.1–0.3% after 7 d of reaction at 50°, and in the range of 3.0–3.9% after 7 d at 90°, independently of the amount of *Celite* used).

Aromatic aldehydes such as [1,1′-biphenyl]-4-carbaldehyde (**4**) were also reductively coupled by H_2S under the same conditions, and GC/MS analysis of the reaction mixture led to the detection of the corresponding (*E*)- and (*Z*)-diarylethenes, **5** and **6**, formed in a 3:1 ratio (*Scheme 2*). Compound **5** was unambiguously identified by comparison of its chromatographic behavior (coinjection experiment using GC) and of its mass spectrum with that of a synthetic standard. However, the main reaction products were di- and trithietiane (based on probe-MS), resulting from di- and trimerization of the thioaldehyde that was formed by sulfurization of the aldehyde **4** [9], as well as the corresponding thiol **7** and the related disulfide **8** (identified by comparison with synthetic standards: mass spectra and coinjection experiments using

Scheme 2. *Formation of Diarylethylenes* **5** *and* **6** ((*E*)/(*Z*) 3:1) *by Reaction of [1,1′-Biphenyl]-4-carbaldehyde* (**4**) *with* H_2S *under Aqueous Conditions*



GC) and trisulfide **9** (identification based on MS data (*m*/*z* 430 (M^+)). The thiol and the di- and trisulfides were likely formed by reduction of the thioaldehyde intermediate **10** following a reductive step which involves single-electron transfer from HS^- [2a,b]. This process competed with the reductive C–C bond formation.

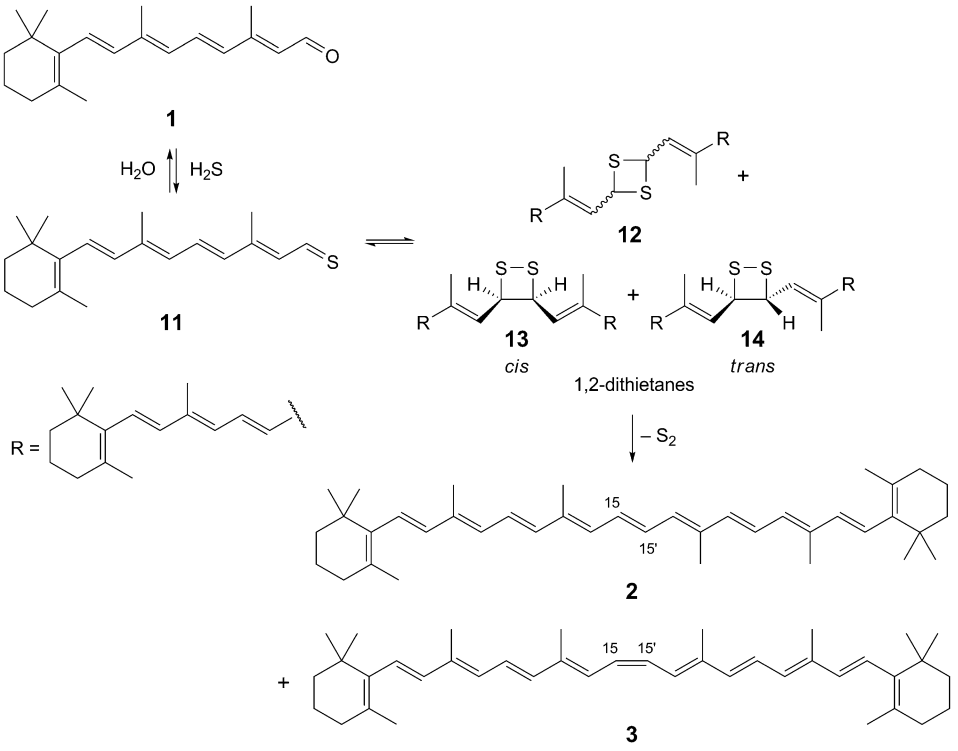
We have envisaged that the formation of intermediate thioaldehydes might play a role in the formation of C–C bonds in our experiments, since thiocarbonyls are known to undergo dimerization under anhydrous conditions with various reagents. Indeed, in the so-called '*Gattermann*' reaction, two thiocarbonyl compounds are reductively coupled to yield substituted ethylenes. This reaction is triggered by various metals such as Cu, Fe, Zn, Bi, Sb, *Raney*-Ni, and *Raney*-Ag, or by a combination of *Lewis* acids and reducing agents such as $FeCl_3/Et_2BH_2Na$, $TiCl_3/K$, and C/Mg [10–13]. These coupling reactions, which are obviously related to the *McMurry* reaction, can only be performed under non-prebiotic (anhydrous) conditions.

However, there are also a few reports of spontaneous coupling of conjugated thiocarbonyl compounds leading to substituted ethylenes without catalysts [14][15]. In the case of thioketones, high temperatures ($>100°$) are required, and these reactions are usually favored by the presence of conjugated electron-withdrawing groups. The reductive coupling of non- or poorly enolizable conjugated aldehydes induced by a thionation reagent (*Lawesson*'s reagent) [16] under anhydrous conditions has also been reported [17]. This process most likely also proceeds *via* the coupling of unstable thiocarbonyl intermediates. It has been proposed that the formation of substituted ethylenes from thiocarbonyl compounds involves the formation of labile 1,2-dithietanes by a [2+2] addition, followed by the elimination of S_2 [15]. This hypothesis was strongly supported by the experiments reported by *Steliou et al.* [18] who developed substrates with two thioaldehyde groups undergoing intramolecular

reductive formation of a C–C bond. The extrusion of S_2 during this process was demonstrated by trapping S_2 with 2,3-diphenylbuta-1,3-diene in a *Diels–Alder* reaction [18]. To the best of our knowledge, there is only one example of stable 1,2-dithiethane (in this case, related to thioesters) reported in the literature [19].

By analogy, we have envisaged that the formation of disubstituted ethylenes from aldehydes under our experimental conditions was the result of the reductive coupling between two thioaldehydes following a process similar to that reported for the thermal coupling of conjugated thiocarbonyls (*Scheme 3*). Indeed, under aqueous conditions and in presence of H_2S, aldehydes might be in equilibrium with small amounts of the related thioaldehydes (*e.g.*, **11**, *Scheme 3*). This equilibrium might ensure a low but constant concentration of thioaldehydes which are available for the coupling reaction, although thioaldehydes are known to be unstable and reactive species which easily polymerize and predominantly occur as 1,3-dithietanes (*e.g.*, **12**; *Scheme 3*) or trithietanes [20][21]. It should be mentioned also, in this respect, that an orange-colored compound characterized by a molecular-ion peak at *m*/*z* 600 (M^+) and a predominant fragment-ion peak at *m*/*z* 300 (probe-MS), possibly corresponding to 1,3-dithietane **12**, could be isolated by reversed-phase HPLC. This compound was, however, too unstable for further NMR study.

Scheme 3. *Formation of 1,2-Dithietane Isomers* **13** *and* **14** *by the Dimerization of Thioretinal* **11** *and Subsequent Generation of β-Carotenes* **2** *and* **3** *by Extrusion of* S_2



Hence, the reductive coupling of the unstable thioaldehyde intermediates might involve a process in which the [2+2] addition, leading to a 1,2-dithietane, is followed by the elimination of S_2. The formation of two possible 1,2-dithietane intermediates **13** and **14** either by *syn* or *anti* [2+2] addition (*Scheme 3*) possibly accounts for the formation of the (*Z*)- and (*E*)-alkenes in our experiments.

A Novel Prebiotic Pathway Leading to the Formation of Compounds from the Citric Acid Cycle. The reductive tricarboxylic acid cycle was postulated to be an autocatalytic cycle which could have played a role in a primordial reductive carbon fixation process [22–24]. In this respect, *Eschenmoser* (2007) [23][24] pointed to a constitutional relationship between glyoxylic acid (**15**), the structurally related HCN dimer (iminoacetonitrile), and various compounds from the reversed reductive citric acid cycle, and proposed that a potentially genetic relationship might even exist between glyoxylic acid and the members of the reductive tricarboxylic acid cycle. This relationship has, however, not been investigated from an experimental point of view up to now. In this context, we have investigated the prebiotic potential of the reductive coupling between non-enolizable aldehydes discussed above, using glyoxylic acid (**15**) as a substrate. Compound **15** is a prebiotically relevant substrate, since it can formally be considered [24] as the dimer of CO and has a non-enolizable aldehyde functionality which might potentially undergo the reductive coupling reaction presented above, leading to fumaric acid (**16**) and maleic acid (**17**). This might potentially open a pathway to the structurally related succinic acid (**18**), malic acid (**19**), and other compounds playing a role in the citric acid cycle.

We have, therefore, performed experiments under conditions similar to those used for the coupling of lipidic aldehydes. Thus, the reaction was carried out in aqueous solution with Na_2S as a reagent. Due to its high H_2O solubility, glyoxylic acid (**15**) was, however, not adsorbed on *Celite* as has been the case when the reaction was performed with lipidic substrates reported above. After 7 d at 90°, the products of the reactions were analyzed (as butyl ester derivatives) by GC and GC/MS (*Fig. 1*). A great variety of compounds was obtained, comprising, notably, fumaric acid (**16**), maleic acid (**17**), succinic acid (**18**), malic acid (**19**), tricarballylic acid (**20**), citric acid (**21**), tartaric acids (**22**/**23**), and hydroxymalonic (tartronic) acid (**24**), which result from the formation of one or two C–C bonds. Oxalic acid (**25**) and compounds resulting from reductive sulfurization [2a,b] of the aldehyde function of glyoxylic acid (*e.g.*, 2-sulfanylacetic acid (**26**; in some of the experiments performed), 2,2′-dithiodiacetic acid (**27**), and related trisulfide **28**) were also evidenced in the reaction mixture. Compounds resulting from both sulfurization and C–C bond forming reactions (*e.g.*, 2-[(carboxymethyl)sulfanyl]succinic acid (**29**) and related disulfide **30**) could also be detected.

The compounds **16**–**29**, except **22** and **28**, have been identified by comparison of their chromatographic behaviors (coinjection experiments using GC) and of their mass spectra with those of commercial or synthetic standards. Structural assignments of compounds **22**, **28**, and **30** are based on the interpretation of mass spectra. In particular, the structures of tartaric acid (*meso*; **22**) and disulfide **30** (as butyl esters) were postulated based on the analogy of their MS-fragmentation pattern with that of DL-tartaric acid (**22**; as butyl ester) and 2-[(carboxymethyl)sulfanyl]succinic acid (**29**; as butyl ester), respectively. The structures of several other compounds occurring in the complex mixture obtained could not be determined comprising compound labeled **X** in

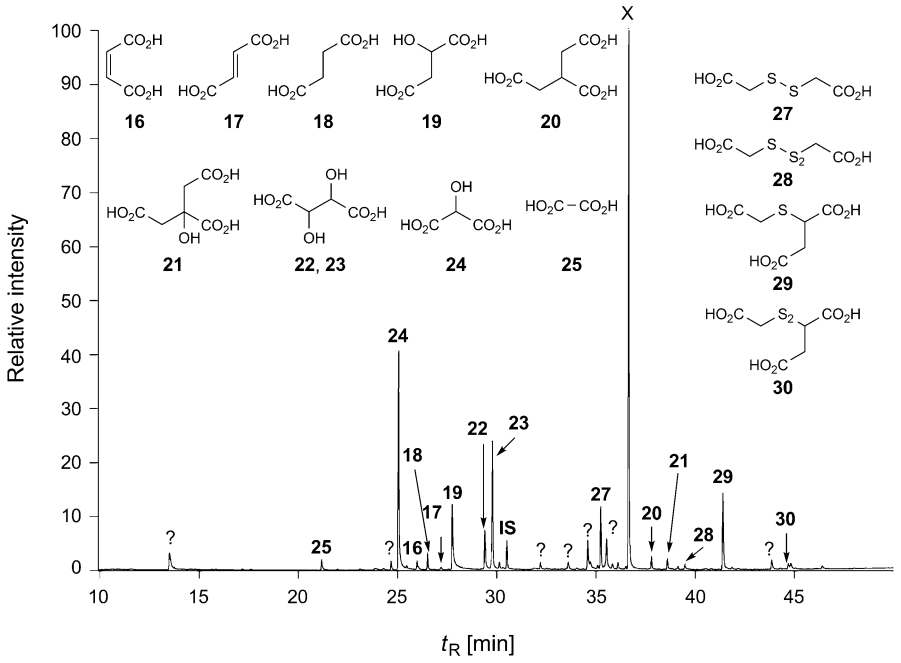


Fig. 1. *Gas chromatogram of the acids* (analyzed as butyl esters) *obtained from the reaction of glyoxylic acid* (**15**) *with* Na_2S *under aqueous conditions*

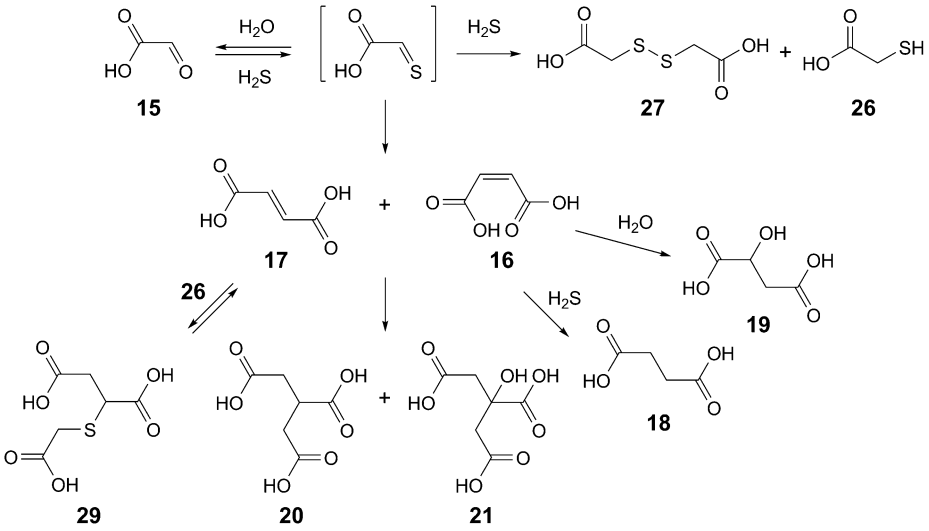
Fig. 1. However, based on HR-MS and on the mass-fragmentation pattern, the formula $C_{17}H_{30}O_7$ was attributed to this compound which most likely corresponds to the butyl ester derivative of a triacid with the formula $C_5H_6O_7$.

The mode of formation of the various products observed and their genetic relationships are depicted in *Scheme 4*. In this respect, the formation of fumaric and maleic acids (**16** and **17**, resp.) can be explained by the novel coupling reaction described in *Scheme 3*. Succinic acid (**18**) likely derives from **16** and **17** by a reductive process possibly induced by $H_2S/NaSH/Na_2S$. The fact that reduced sulfur species are able to reduce various organic functionalities, comprising conjugated C=C bonds, is indeed well-documented [2d]. Malic acid (**19**) and 2-[(carboxymethyl)sulfanyl]succinic acid (**29**) result most likely from the *Michael*-type addition of H_2O and 2-sulfanylacetic acid (**26**) on fumaric or maleic acid, respectively.

2-Sulfanylacetic acid (**26**) and 2,2′-dithiodiacetic acid (**27**) are formed by reductive sulfurization of the carbonyl functionality of glyoxylic acid induced by sulfides [2a,b]. This is a well-established reaction occurring in natural environments, and affecting ketones and aldehydes in anoxic sedimentary settings where intense sulfate reductions occur [2a,b].

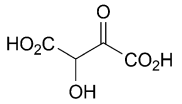
The formation of citric acid (**21**) and tricarballylic acid (**20**), which necessarily implies the coupling of three glyoxylic acid units, followed by reductive processes most likely involving reduced sulfur species [2], could not be fully elucidated.

Scheme 4. *Genetic Relationships between the Identified Products Formed by Reaction of Glyoxylic Acid* (**15**) *with* Na_2S *under Aqueous Conditions*



Quantification of the identified products obtained showed that the sum of fumaric, maleic, succinic, malic, tricarballylic, citric acids (**16**–**21**, resp.), 2-[(carboxymethyl)-sulfanyl]succinic acid (**29**), and related disulfide **30**, resulting from C–C bond formation and genetically related to **16** and **17** (the compounds primarily formed from glyoxylic acid (**15**) following the new reductive coupling reaction) represents 18% of **15** used as substrate.

The presence of tartaric acid isomers **22** and **23** might possibly be explained by another C–C bond-forming process operative in parallel to that presented above. Tartaric acids **22** and **23** might be formed by the coupling of glyoxylic acid (**15**) by a reductive process related to the pinacolic coupling [25] or by a process related to the benzoin condensation (possibly involving *Umpolung via* dithioacetal-type structures) which would yield oxaloglycolic acid (**31**) as an intermediate. Further degradation of **31** might also explain the formation of tartronic acid (**24**) [26]. Investigation on the possible formation of oxaloglycolic acid (**31**) as intermediate is currently underway.



Oxaloglycolic acid (**31**)

Formation of C–C Bond by Pyrolysis of 2,2′-Dithiodiacetic Acid (**27**). A high-temperature pyrolysis of Me_2S_2 has previously been reported to yield ethylene [27]. We have, therefore, envisaged that disulfides, like compound **27**, which are important products formed by reaction of aldehydes with H_2S under aqueous conditions [2a,b] might further lead to C–C bond formation provided that temperature conditions permitting S–S bond cleavage are attained. This question is also relevant with respect to chemical reactions of prebiotic interest likely to occur in the context of hydrothermal

vents where sulfide-rich fluids are emitted in the seawater at high temperature [4]. We have, therefore, performed an experiment where 2,2′-dithiodiacetic acid (**27**) was heated in a sealed tube for 4 h at 200°. We observed, among the products formed, succinic acid (**18**), carballylic acid (**20**), as well as 2-[(carboxymethyl)sulfanyl]succinic acid (**29**) and the related disulfide **30** as major compounds (*Fig. 2*).

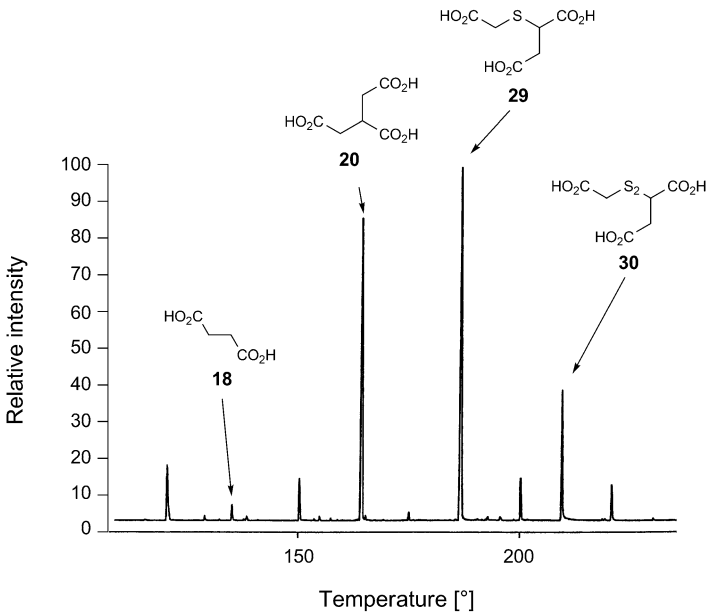


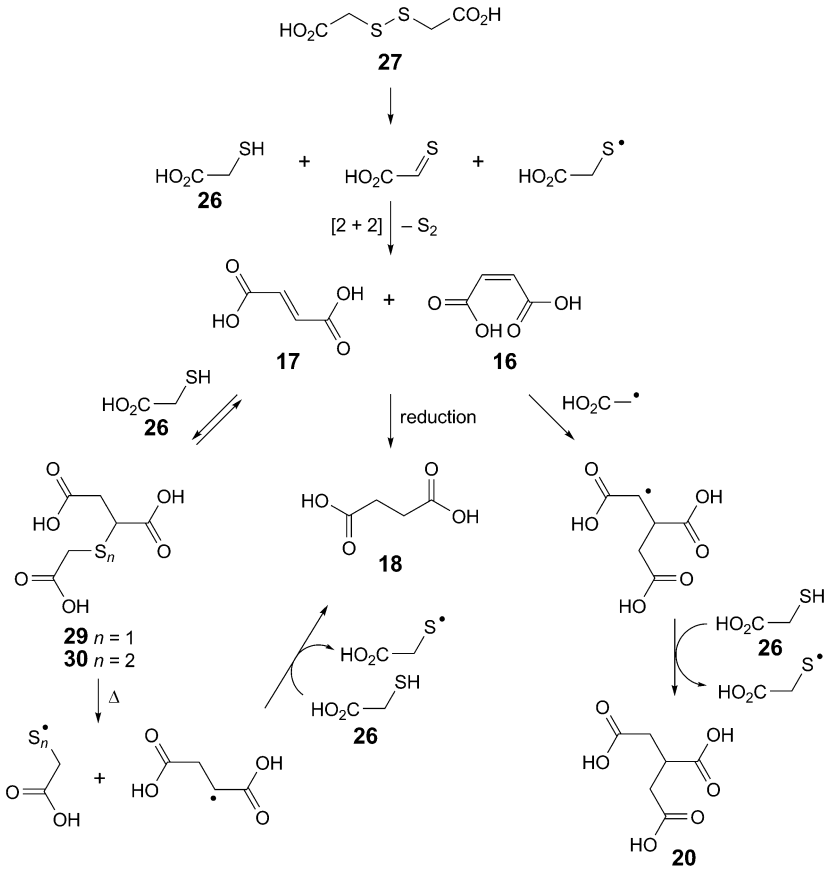
Fig. 2. *Gas chromatogram of the acids* (analyzed as butyl esters) *obtained from thermal treatment* (200°, 24 h) *of 2,2′-dithiodiacetic acid* (**27**)

Pyrolysis of disufide **27** most likely results, in a first step, in the formation of a thiocarbonyl intermediate (as has been described for Me_2S_2 [27]). Reductive coupling of this thiocarbonyl compound might then lead to the formation of fumaric acid (**16**) and maleic acid (**17**) (*Scheme 5*). Possible pathways leading to 2-[(carboxymethyl)-sulfanyl]succinic acids (**29**), succinic acid (**18**), and carballylic acid (**20**) are described in *Scheme 5*.

The results obtained from the reported pyrolysis experiment clearly show that disulfides (like compound **27**) formed by reductive sulfurization of aldehydes, induced by H_2S, HS^-, or S^{2-} ions [2a,b], are not end products and can further react to form C–C bonds under high-temperature conditions. This is relevant in the context of hydrothermal vents.

Conclusions. – We have shown that non- or poorly enolizable aldehydes can undergo a dimerization reaction in H_2O in the presence of H_2S or S^{2-} ions to yield alkenes *via* a reductive coupling reaction, which involves a [2+2] addition of intermediate thioaldehydes, followed by a *retro*-[2+2] extrusion of S_2. This coupling reaction is in competition with two other major reactions: the oligomerization of the thioaldehydes leading to 1,3-dithietanes or trithietanes, and the reduction of the

Scheme 5. *Possible Mode of Formation of the Main Products Formed upon Thermal Treatment* (200°, 24 h) *of 2,2′-Dithiodiacetic Acid* (**27**)



thiocarbonyl compounds into the corresponding thiols or disulfides. These side reactions, along with the low concentration of the thioaldehydes in aqueous medium, can account for the low yield of the reductive coupling reaction observed in the case of lipidic substrates. This reaction, performed in aqueous medium, has clear relevance to prebiotic chemistry. In this respect, when glyoxylic acid (**15**) was used as substrate, various compounds from the reversed citric acid cycle (fumaric, succinic, malic, and citric acids) or of related compounds (carballylic acid) were formed. The reversed citric acid cycle was indeed postulated to have played a role in a primordial reductive carbon fixation process [22–24]. Such C–C bond-forming reactions might, notably, have been operative at the sulfide-rich environments of hydrothermal vents, which have been postulated as possible sites for the first steps of organic chemical evolution [28].

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Experimental Part

1. *General.* All solvents were distilled before use. Flash chromatography (FC) and column chromatography (CC): silica gel *60* (SiO_2; 40–63 μm, *Merck*). NMR Spectra: *Bruker Avance 300* (^1H: 300 and ^{13}C: 75 MHz), *Bruker AM 400* spectrometer (^1H: 400 and ^{13}C: 100 MHz), or *Bruker ARX 500* (^1H: 500 MHz and ^{13}C: 125 MHz) with CD_2Cl_2 or C_6D_6 as solvents; δ in ppm rel. to Me_4Si as internal standard, J in Hz.

GC Analyses were carried out on a *HP 6890* gas chromatograph equipped with an on-column injector, a FID (flame ionization detector; 300°) and a *HP-5* MS column (30 m × 0.32 mm i.d., 0.25-μm film thickness). H_2 was used as carrier gas (2.5 ml/min). The temp. programs were: 40° (5 min); 40–300° (10°/min); 300°, isothermal or 40–100° (10°/min); 100–300° (4°/min); 300°, isothermal. GC/MS Analyses were carried out either on a *Finnigan MAT TSQ 700* mass spectrometer connected to a *Varian 3400* gas chromatograph equipped with an on-column injector and with a *RTX5*-MS column (60 m × 0.32 mm i.d., 0.1-μm film thickness), on a *Varian 1200L* mass spectrometer connected to a V*arian CP 3800* gas chromatograph equipped with an on-column injector and with a *HP-5* MS column (30 m × 0.32 mm i.d., 0.25 μm film thickness), or on a *Thermo Scientific TSQ* quantum spectrometer connected to a *Trace* gas chromatograph (PTV injector), and with a *HP-5* MS column (30 m × 0.32 mm i.d., 0.25-μm film thickness). MS: either electronic impact (EI) ionization at 70 eV or chemical ionization (CI) with isobutane or NH_3 as ionization gas; He was used as carrier gas.

2. *Reductive Coupling of Conjugated Aldehydes. Reductive Coupling of Lipidic Substrates.* A soln. of substrate (retinal (**1**) or arom. aldehyde **4**; 20 mg) was added to a large excess of *Celite* (7 g). The solvent was removed under reduced pressure insuring a good dispersion of the substrate on the *Celite*. The solid phase obtained was suspended in a 0.1M aq. NaOH soln. (50 ml), and the suspension was saturated with H_2S. After heating at 50° or 90° for periods extending from 7 to 15 d, the soln. was degassed under a flow of Ar, and the *Celite* was recovered by filtration. The crude mixture obtained by extraction of the *Celite* with CH_2Cl_2 was fractionated on a SiO_2 column (hexane/CH_2Cl_2 8:2) to yield a non-polar fraction containing the hydrocarbons and the org. sulfur compounds, and a more polar fraction containing the residual substrate. The fractions obtained were analyzed by probe-MS, GC/MS and, in the experiments performed on retinal (**1**), by HPLC/UV/VIS (*DuPont Zorbax ODS* 250 × 4.6 mm, 5 μm, eluent acetone/MeOH 1:1, 1 ml/min). β-Carotene (**2**) was isolated from the mixture by HPLC using the conditions described above. In the case of the experiments with **1**, a 20-ml soln. in EtOH of the non-polar fraction isolated from the crude mixture according to the procedure described above was used for quant. analysis of β-carotene (**2**; see *Sect. 4*).

Reductive Coupling of Glyoxylic Acid (**15**). A mixture of **15** · H_2O (40 mg, 0.43 mmol) and Na_2S (as nonahydrate; 480 mg, 2.00 mmol) in dist. degassed H_2O (3 ml) was heated at 90° for typically 7 d. At the end of the reaction, adipic acid was added as a standard for quantification, and the soln. was evaporated to dryness under reduced pressure. The acids were transformed into the related butyl esters prior GC/MS analysis by treatment of the dry residue with a 1.5M soln. of HCl in BuOH (90° for 2 h). The products were identified by GC/MS and were compared with commercial and synthetic standards (see *Sect. 3*).

Pyrolysis of Disulfides. In a typical experiment, *2,2′-dithiodiacetic acid* (**27**; 20 mg) was sealed in a glass tube under vacuum and heated at 200° for 4 h. The crude mixture obtained was treated with BuOH/HCl according to the procedure described above and analyzed by GC/MS. The products were identified by GC/MS and were compared with commercial and synthetic standards (see *Sect. 3*).

3. *Characterization of Compounds Formed in Laboratory Experiments.* The different compounds formed during the laboratory experiments involving **15** or **27** as substrate were identified by interpretation of the mass spectra obtained using EI and CI GC/MS. The structures of some key products were established by comparison of MS data and chromatographic behavior (co-elution experiments using GC) with those of reference standards obtained by synthesis (*tartronic acid* (**24**), *2-[(carboxymethyl)sulfanyl]succinic acid* (**29**); see *Sect. 6*) or from commercially available compounds after esterification with BuOH/HCl (*fumaric acid* (**16**), *maleic acid* (**17**), *succinic acid* (**18**), *malic acid* (**19**), *carballylic acid* (**20**), *citric acid* (**21**), DL-*tartaric acid* (**23**), *oxalic acid* (**25**), *2-sulfanylacetic acid* (**26**), and **27**).

In the case of the experiments on **1**, β-carotene identification was based on the comparison of the ^1H-NMR spectrum (C_6D_6) of isolated *β-carotene* (**2**) formed from reductive coupling of retinal (**1**) and of commercial β-carotene (**2**) which proved to be almost superimposable.

In the case of the experiment involving *[1,1′-biphenyl]-4-carbaldehyde* (**4**) as substrate, *(*E*)-1,2-di([1,1′-biphenyl-4-yl])ethene* (**5**), *[1,1′-biphenyl]-4-methanethiol* (**7**), and *1,2-bis([1,1′-biphenyl]-4-ylmethyl)disulfane* (**8**) were identified by comparison (MS and t_R in GC, co-elution experiments) with reference standards obtained by synthesis (see *Sect. 6*).

4. *Quant. Analysis of β-Carotene* (**2**) *Formed by Reductive Coupling of* **1**. Quantification of β-carotene (**2**) was accomplished by HPLC coupled with a PDA detector (*Waters 996*) using a *DuPont Zorbax ODS* column (250 × 4.6 mm, 5 μm; acetone/MeOH 1:1, 1 ml/min). A calibration curve was established using solns. prepared with various concentrations of **2** (0.1–0.8M). The solns. (in EtOH) of the apolar fractions containing **2** obtained by fractionation of the crude mixture after the experiments (see *Sect. 2*) were used for quant. analyses by HPLC. The equation of the calibration curve was then used to calculate the concentrations of **2** formed.

5. *Quant. Analysis of Products Formed in the Experiments with* **15**. The yields of the identified compounds resulting from a process involving the reductive coupling of aldehydes were determined in the experiment involving **15** (*cf. Fig. 1*). Quantification was accomplished by integration on the GC-FID chromatograms and by comparing the area under the peaks of the different compounds (as dibutyl esters) observed with the area of the peak of adipic acid (as dibutyl ester) added as quantification standard. The response factors have not been experimentally determined and were assumed to be proportional to the number of C-atoms.

We determined the proportion relative to **15** used as substrate of C_2 moieties (related to **15**) occurring in C_4 and C_6 compounds genetically related to **16** and **17** (*Scheme 4*). This proportion could be considered as an overall yield of the C–C bond-forming reactions relative to **15**. The calculation of this proportion is based on the quant. data obtained for the following compounds: **16**–**21**, **29**, and disulfide **30**.

6. *Synthesis of Reference Standards. Synthesis of* **5**. [(1,1′-Biphenyl)-4-methyl]triphenylphosphonium chloride (2.00 g, 4.30 mmol) was dissolved in 30 ml of THF. BuLi (0.65N, 7.90 ml, 5.16 mmol, 1.2 equiv.) was added under Ar to give a red soln. After 30 min, (1,1′-biphenyl)-4-carbaldehyde (1.18 g, 6.45 mmol, 1.5 equiv.), dissolved in 10 ml of THF, is added leading to an instantaneous discoloration of the soln. The mixture was poured in dist. H_2O and extracted (3 ×) with CH_2Cl_2. Purification of the crude mixture by CC (SiO_2; hexane/CH_2Cl_2 4:1) yielded **5** (1.42 g, 4.28 mmol). ^{1}H-NMR (300 MHz, C_6D_6): 6.23 (*s*, 2 H); 7.14–7.28 (*m*, arom. H); 7.40–7.52 (*m*, 4 arom. H). ^{13}C-NMR (75 MHz, C_6D_6): 127.0; 127.1; 127.2; 128.8; 129.6; 130.0; 135.6; 140.2; 140.8. GC/EI-MS: 332 (100, M^+), 330 (2), 317 (15), 252 (11), 241 (16), 239 (11), 178 (3), 176 (3), 166 (6).

Synthesis of **7**. 4-(Chloromethyl)[1,1′-biphenyl] (3.01 g, 14.9 mmol) was dissolved in 40 ml of DMF under Ar. AcSK (1.80 g, 15.9 mmol, 1.06 equiv.) was added, and the mixture was stirred for 1 h. The mixture was then poured in dist. H_2O and extracted (4 ×) with hexane. Purification of the crude mixture by CC (SiO_2; hexane/CH_2Cl_2 1:1) led to [1,1′-biphenyl]-4-methylsulfanyl acetate (3.56 g; 14.7 mmol) in 98% yield. The related thiol **7** was obtained by treatment of the acetate with KOH/MeOH in quant. yield.

Data of (1,1′-Biphenyl)-4-methylsulfanyl Acetate. ^{1}H-NMR (300 MHz, C_6D_6): 1.89 (*s*, Ac); 4.07 (*s*, PhCH_2–SAc); 7.16–7.28 (*m*, arom. H), 7.40–7.49 (*m*, 4 arom. H). ^{13}C-NMR (75 MHz; C_6D_6): 29.5; 33.0; 127.1; 127.2; 127.4; 128.8; 129.4; 137.1; 140.4; 141.0; 193.5. GC/EI-MS: 242 (19, M^+), 199 (3), 167 (100), 165 (22), 152 (11), 115 (7), 89 (2).

Data of **7**. ^{1}H-NMR (300 MHz; C_6D_6): 1.53 (*t*, J = 7.5, CH_2SH); 3.38 (*d*, J = 7.5, CH_2SH); 7.12–7.31 (*m*, arom. H); 7.42–7.53 (*m*, 4 arom. H). ^{13}C-NMR (75 MHz; C_6D_6): 28.4; 127.1; 127.2; 127.3; 128.5; 128.8; 140.0; 140.3; 141.0. GC/EI-MS: 200 (21, M^+), 167 (100), 165 (25), 152 (15), 115 (3), 82 (7).

Synthesis of **8**. Compound **7** (700 mg, 3.49 mmol) was dissolved under Ar in 150 ml of MeOH/EtOH 2:1. Cat. amounts of Na_2CO_3 and I_2 (440 mg, 1.73 mmol), dissolved in 5 ml of MeOH, were added, and the mixture was stirred for 30 min. The solvent was then removed under reduced pressure. The solid residue obtained was dissolved in CH_2Cl_2, and the org. phase was washed with dist. H_2O (3 ×). Purification of the crude mixture by CC (SiO_2; hexane/CH_2Cl_2 1:1) yielded **8** (696 mg; 1.74 mmol). ^{1}H-NMR (300 MHz; C_6D_6): 3.56 (*s*, 2 PhCH_2S); 7.16–7.30 (*m*, arom. H); 7.45–7.52 (*m*, arom. H). ^{13}C-NMR (75 MHz; C_6D_6): 42.9; 127.1; 127.2; 127.3; 128.8; 130.0; 135.6; 140.5; 140.9. GC/EI-MS: 398 (7, M^+), 199 (3), 167 (100), 165 (42), 152 (15), 115 (2), 89 (1).

Synthesis of 2-Hydroxypropanedioic Acid (= *Tartronic Acid*; **24**). Mesoxalic acid monohydrate disodium salt (50 mg, 0.28 mmol) and $NaBH_4$ (53 mg, 1.40 mmol) were reacted in 1.5 ml of dist. H_2O. After 2 h, the soln. was acidified with an aq. soln. of HCl. The resulting mixture was evaporated to dryness under reduced pressure, and the dry residue was treated with a 1.5M soln. of HCl in BuOH (90° for 2 h). The residue obtained after evaporation of BuOH was purified by FC (SiO_2; AcOEt). Compound **24** was obtained as dibutyl ester in almost quant. yield. ^{1}H-NMR (400 MHz; CD_2Cl_2): 0.93 (*dd*, *J* = 7.5, 7.5, 6 H); 1.37 (*m*, 4 H); 1.64 (*m*, 4 H); 4.20 (*m*, 4 H); 4.67 (*s*, C*H*OH). ^{13}C-NMR (100 MHz; CD_2Cl_2): 13.3; 18.9; 30.4; 66.3; 71.5; 168.7. GC/EI-MS (70 eV, rel. int.): 132 (22), 121 (9), 76 (100), 57 (90). GC/CI-MS: 250 (100, $[M+NH_4]^+$), 233 (40, $[M+H]^+$).

Synthesis of **29**. Compound **26** (32 mg, 0.35 mmol) was added to **16** (40 mg, 0.35 mmol) and NaOH (70 mg, 1.75 mmol) in 5 ml of dist. degassed H_2O. The mixture was heated at 90° for 2 d. The resulting mixture was evaporated to dryness under reduced pressure. The acids were transformed into the corresponding butyl esters by treatment of the dry residue with a 1.5M soln. of HCl in BuOH (90° for 2 h). The residue obtained after evaporation of BuOH was purified by FC (SiO_2; CH_2Cl_2/cyclohexane 9:1). Compound **29** was obtained as butyl ester in almost quant. yield. ^{1}H-NMR (400 MHz; CD_2Cl_2): 0.94 (*dd*, *J* = 7.4, 7.4, 9 H); 1.38 (*s*, 6 H); 1.62 (*s*, 2 Me); 2.69 (*dd*, *J* = 16.5, 5.0, 1 H); 2.94 (*dd*, *J* = 16.5, 10.1, 1 H); 3.35 (*d*, *J* = 15.6, 1 H); 3.51 (*d*, *J* = 15.6, 1 H); 3.80 (*dd*, *J* = 10.1, 5.2, 1 H); 4.07 (*m*, 6 H). ^{13}C-NMR (100 MHz; CD_2Cl_2): 13.4; 19.0; 30.5; 33.3; 36.2; 41.8; 65.5; 169.6; 170.3; 170.5. GC/EI-MS: 376 (40, M^+), 302 (93), 274 (52), 246 (91), 230 (76), 218 (69), 200 (92), 190 (82), 175 (88), 163 (63), 156 (75), 144 (100), 131(60), 119 (45), 100 (94), 73 (39), 57 (94). HR-ESI-MS: 399.1813 ($[M+Na]^+$, $C_{18}H_{32}NaO_6S^+$; calc. 399.1817).

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