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Enantioselective reactions catalyzed by synthetic enzymes. A model for chemical evolution

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ABSTRACT

Polyleucines of various lengths act as enantioselective catalysts in the aldol condensation between cyclohexanone and various aromatic aldehydes. Polyleucine and other polyamino acids behave as synthetic enzymes in the epoxidation of chalcone and other electron-deficient alkenes. Both reactions are of considerable prebiotic significance.

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1. Introduction

As shown by numerous experiments that have simulated a wide range of primitive environments, there is strong support for the hypothesis that the origin of life was preceded by the synthesis and accumulation of α-amino acids, nucleobases, and many other organic compounds of biochemical significance.

The inventory of prebiotic catalytic agents may have included metallic cations, clays, imidazole derivatives, highly reactive derivatives of HCN (cyanamide and dicyanamide and cyanogens), as well as chemically active small peptides and oligopeptides.

In this largely accepted scenario, recently developed experimental systems have clarified the possible in vitro functional evolution of RNA,[1] and the template-directed synthesis of a genetic polymer in model protocells.[2] Together with the reported syntheses of all the relevant precursors in abiotically plausible conditions,[3,4] their non-enzymatic activation,[5] and the synthesis of activated pyrimidine ribonucleotides,[6] these observations hint to the possible solution of the origin of polymeric (pre)-genetic information.

Although the synthesis and the polymerization of nucleobases have long lagged behind the abiotic synthesis of amino acids, as reported in the seminal Miller experiments[7] and in recent reports,[8,9] the answer to the origin of nucleic acids seems to be more at hand than that of proteins. In this respect, two reports[7,10] have critically evaluated the possibility of oligomerization of glycine, pointing to the difficulties encountered when trying to establish the conditions for the abiotic synthesis of even the simpler oligopeptides.

The self-sustaining replication systems that characterize extant life are based on both nucleic acids and proteins. Thus, even an initial understanding of the events that kick-started replication and evolution requires that oligomerization of peptides is brought into a thermodynamically sound frame of reference and that the problem of chirality, that typically characterizes amino acids when considered as biological precursors, is eventually solved.

The emergence of homochirality in the prebiotic world is on the one hand a major open problem, while on the other hand it provides a fascinating challenge for investigating biomimetic catalysis. In recent years, significant progress has been made in the design of synthetic polypeptides, which are able to catalyze chemical reactions similar to those promoted by enzymes, although generally with significantly lower efficiency.

The peptide part of the scheme generally accepted for the origin of life, consists of four fundamental steps: (i) the abiotic formation

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of α-amino acids; (ii) symmetry breaking leading to chiral building blocks having small enantiomeric excess (ee); (iii) the chiral amplification to enantiopure substances; and (iv) their organization into self-sustaining systems.

Attempts to explain the origin of biological homochirality have led to a large variety of proposals.[11] Most of them assume the preferential destruction of one of the enantiomers, which may have accumulated as racemic mixtures in the prebiotic environment as a result of abiotic synthesis.

Recent experiments suggest that some of the catalytic species mentioned in the inventory reported above, from metallic cations to imidazole derivatives and to chemically active small peptides, may have played a central role in prebiotic enantioselective synthesis.

In this context, Reza Ghadiri et al. have recently shown that carbonyl sulfide, a simple volcanic gas, is able to form peptides from simple α amino acids in aqueous solution and under very mild conditions in yields up to 80% in minutes to hours at room temperature.[12] The same authors have also found that this gas, a constituent not only of volcanic gas emission on Earth, but also of the atmosphere of Venus and present in interstellar gas clouds, could have mediated, under mild aqueous conditions, both the phosphoryl transfer and the peptide synthesis from aminoacyl *N*-carboxy-anhydride as a single intermediate on prebiotic Earth.[13] Finally, Ghadiri described a supramolecular peptide assembly that catalyzes diketopiperazine and dipeptide synthesis from a variety of aminoacyl substrates.[14]

Asymmetric organocatalysis has become, in the last few years, one of the most active fields of research in organic chemistry, since it is based on the ability of small organic molecules to catalyze reactions in a stereoselective way with ease. A large number of powerful asymmetric bond-forming reactions have been reported, that allow for the enantioselective synthesis of a great variety of compounds. In 2002, Barbas prepared carbohydrates, polyketides, and unusual amino acids using a very versatile tool, namely the organocatalytic asymmetric aldol, Mannich, Diels-Alder, and other reactions, proposing that organocatalysis could have been involved in the prebiotic synthesis of the building blocks of life.[15] The work of Blackmond, Hayashi, and Breslow[16–19] has opened up the route for obtaining nearly enantiomerically pure amino acids from only slightly enantiomerically enriched mixtures. This mechanism might act as a propelling machinery for producing the homochirality of the prebiotic world.

The same considerations apply to peptides as chiral catalysts. Indeed, as shown by the biologically active random co-polymers of glutamate and phenylalanine[20,21] and the catalytic activity of histidyl-histidine,[22] structurally simple polyamino acids of abiotic origin may have exhibited catalytic activities possibly leading to asymmetric synthesis.

The role of (*R*)-His-(*R*)-Phe in catalyzing the addition of HCN to aldehydes to give optically active cyanohydrins demonstrates the potential of the use of simple peptides as catalysts in an asymmetric system with great prebiotic relevance.[23]

2. Aldol condensation

The aldol reaction, is a key reaction in organic synthesis, with considerable biological interest since it forms C–C bonds.[24] List, Lerner, and Barbas performed the first milestone experiments in polar organic solvents. Since then many asymmetric aldol reactions, via organocatalysis with α amino acids, have been reported.[25] The thermodynamic control of asymmetric amplification, based on the equilibrium solid–liquid phase behavior of amino acids in solution, increases the synthetic value of this procedure.[26]

The aldol reaction is one of the most powerful tools for the formation of C–C bonds in organic synthesis,[27] since it provides enantiomerically enriched beta-hydroxy carbonyl compounds. Simple amino acids show the marvelous efficiency of aldolase enzymes, without the structural complexity required by this class of enzymes. Among the other asymmetric catalytic methods, the proline-catalyzed version, which proceeds via an in situ-generated enamine intermediate, has attracted much attention, leading to products with high chemo- and stereo-selectivity under very mild conditions.[28] In addition to the natural amino acid itself, various proline derivatives and many other small molecules have been successfully used as catalysts.[29,30]

Simple α-amino acids catalyze not only intramolecular but also intermolecular aldol reactions with high enantioselectivity and high yield not only in organic solvent reaction media[16,26,31] but also in water solution,[32] a solvent compatible with a prebiotic scenario of the origin of life.[31] The catalytic asymmetric effect of non-racemic alanine and valine in a water-based prebiotic carbohydrate synthesis from glycoaldehyde and formaldehyde has been mentioned in this context,[33] along with the efficient peptide-catalyzed stereospecific synthesis of tetroses by aldol condensation of glycolaldehyde.[34] In addition, Cordova et al. have recently reported that small peptides catalyze asymmetric condensations with up to 99% ee, that is, with stereoselectivities comparable with those of natural enzymes.[35]

Some of us have recently studied the organo-catalyzed cross aldol reaction of cyclohexanone and aromatic aldehydes promoted by submolar quantities of alanine, leucine, and several leucine oligomers of different lengths, in DMSO/H_2O as a solvent. The 4-chlorobenzaldehyde was chosen as the model substrate, and the reaction with four other aromatic aldehydes has also been investigated[36] (Scheme 1).

Anti-Aldols were formed preferentially in all cases with the enantioselectivity being higher for the leucine-catalyzed reaction, in contrast with the lower values observed for the numerous polyleucines analyzed. The best results in terms of chemical yield, enantio- and diastereoselectivities, dr *anti*/*syn* 7:1; ee *anti* 65%, respectively, were obtained with PLL-Bayer, containing 7–8 monomers on average.

The prebiotic significance of aldehydes in the synthesis of sugars, amino acids, hydroxy acids, and other organic molecules in potential biochemical reactions is well established.[37]

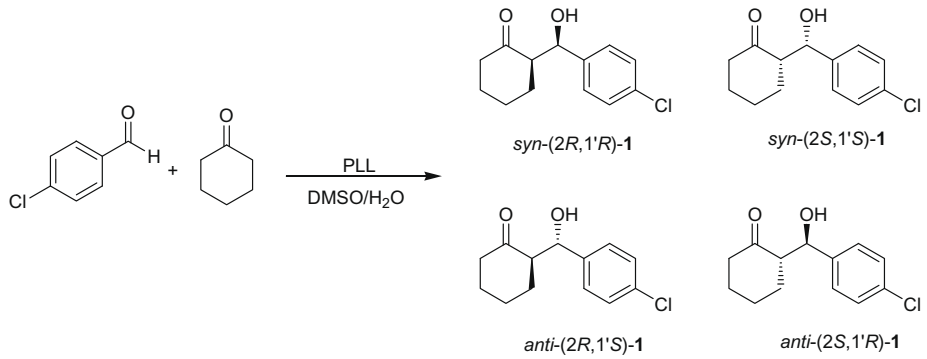
In order to gain further information on the catalytic role of small peptides in reactions involving keto-bearing substrates, other related aromatic aldehydes have been investigated. Moderate to high enantio- and diastereoselectivities were obtained with simple L-leucine as a catalyst, whereas with PLL-Bayer, the enantio- and diastereosectivities were of the same order of magnitude of those observed with the model substrate.

In all cases the prevailing *anti*-diastereoisomer was formed. With α-methyl cinnamaldeyde the results with PLL-Bayer, (yield and enantioselectivity) were comparable or even higher (diastereoselectivity), than those obtained with leucine.

We are aware that the formation of large polypeptides from the same amino acid in the prebiotic environment is unlikely. Testing the possibility of chiral amplification by lengthy peptides was nevertheless of interest. Thus, the role of scalemic polyleucines was explored. In contrast with the amplification effects observed in asymmetric epoxidation of chalcone with hydrogen peroxide in alkaline medium[38] the effect of PLL on the aldol condensation of cyclohexanone with 4-chlorobenzaldehyde was negligible. This may be due to the higher degree of polymerization of the catalyst, consisting of 64 monomers on average.

Non-linear effects in the acyclic amino acid-catalyzed aldol condensation of cyclohexanone with 4-nitrobenzaldehyde in heterogeneous wet DMSO have recently been reported,[39] which could have possible implications in the evolution of homochirality.

The prebiotic availability of the reactants used in our model experiments may be questioned. The DMSO/water medium used by us and by others,[31] may also not be compatible with a prebiotic



Scheme 1. Aldol condensation of 4-substituted benzaldehyde with cyclohexanone.

scenario. However, the significance of these results as a possible route to homochirality is validated by recent reports on aldol condensation in aqueous medium devoid of organic solvents,[32] thus stressing the potential of simple amino acids and of polyleucines in enantioselective intermolecular aldol condensation reactions.

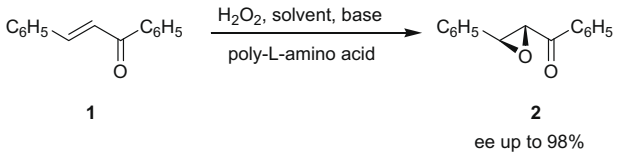
3. Enantioselective epoxidation

Another seminal reaction in organic chemistry is the epoxidation reaction. An elegant and very versatile method for the preparation of enantiomerically pure epoxides continues to be one of the most exciting fields of asymmetric catalysis, due to the high reactivity and versatility of these compounds as chiral synthons. Current methods for the catalytic asymmetric oxidation of electron rich olefins are largely based on the use of chiral metal complexes[40] or on the use of organocatalyst such as chiral ketones.[41]

On the other hand, no systems for the asymmetric epoxidation of electron-deficient olefins have gained widespread popularity as a synthetic tool. Indeed, only recently have systems been described for the epoxidation of a wide range of enones with high enantioselectivity, thus allowing us to take advantage of the potential of epoxides substituted with electron-withdrawing groups as synthetic intermediates.[42] The poly-α amino acid-catalyzed asymmetric Julià-Colonna epoxidation has emerged as an efficient synthetic method for the enantioselective functionalization of α,β-unsaturated ketones. Since its discovery in 1980, the reaction conditions (see later) have been constantly optimized.

Our original procedure[43–45] for the epoxidation of electron poor olefins was based on a triphasic system (polyamino acid/aqueous phase/organic phase) (Scheme 1).

Initially, three polyamino acids, poly-L-alanine, poly-L-α-benzylglutamate, and poly-L-α-butylglutamate, were tested for the oxidation of (*E*)-chalcone **1** in a triphase system in the presence of hydrogen peroxide, organic solvent, and a base (Scheme 2). Successively, the study was extended to a much broader series of amino acids.[44] Ala, Leu, and Ile were the most efficient catalysts and higher polymer homologues compared favorably, in terms of reaction rates and enantiomeric excesses (ee), with the shorter ones. As expected, D-amino acids provided epoxides with the opposite absolute stereochemistry (Table 1).



Scheme 2. Poly-L-amino acid-catalyzed oxidation of chalcone.

Table 1
Epoxidation of chalcone **1** with various polyamino acids

Entry	Catalyst (MW)	Time (h)	Yield (%)	ee%
1	L-Ala (~1000)	28	75	93
2	D-Ala (~1000)	37	53	90
3	D-,L-Ala (~1000)	24	5	0
4	L-Ala (~3000)	28	77	96
5	L-Val (~1000)	168	6	10
6	L-Val (~3000)	144	4	33
7	L-Leu (~1500)	28	60	84
8	L-Leu (~4500)	28	44	88
9	L-Isoleu (~1500)	72	76	95
10	L-Phe (~2000)	72	32	1
11	L-β-Bn-Asp	456	8	3
12	L-β-Bn-Glu	144	12	12
13	[L-Leu-L-Ala]$_{10}$	24	67	95
14	[L-Val-L-Ala]$_{10}$	96	39	88
15	L-Ala-L-Ala	144	41	2

The polyamino acids still functioned as catalysts when anchored on a polystyrene matrix, giving high yield and ee. Substituted chalcones afforded satisfactory results under the standard conditions. Despite the fact that the methodology of the Julià-Colonna reaction has been successfully applied by other authors to a relatively wide range of different substrates, a number of drawbacks limit the applicability of this procedure, especially the long reaction time and the sensitivity of many α,β-unsaturated ketones to the basic nucleophilic aqueous three-phase conditions. Moreover, the work-up of the reactions was rather troublesome in connection with the separation of the highly swollen polyaminoacid gel and it was often accompanied by a decrease in the enantioselectivity when the catalyst was recycled.

The limitations mentioned above have been overcome employing two new methodologies: (i) using water-free conditions as introduced by Robers et al.,[45] and (ii) supporting polyamino acids onto organic[46] or inorganic materials.[47]

In the former case, fundamental enhancement was provided by the use of a urea-hydrogen peroxide complex (UHP), together with an organic base such as 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU), in a non-aqueous two-phase system.[48] Under these conditions the rate of epoxidation of the model substrate chalcone **1** was increased by two orders of magnitude and a larger number of substituted enones were able to react, in a short reaction time, with excellent yield (90–98%) and enantioselectivity (97–99%).

In the latter approach, the polyaminoacid catalysts were immobilized onto cross-linked aminomethyl-polystyrene (85% yield, 99% ee) or silica (93% ee). In both cases the catalyst is easily prepared, more active, and the work-up procedure is much easier.

Recently a soluble version of the Julià-Colonna catalyst has been developed using aminopoly(ethyleneglycol) (PEG-NH_2) to initiate

the polymerization of *N*-carboxyanhydride precursors.[49,50] The resulting peptide conjugate is soluble in THF and allowed the stereoselective oxidation of chalcone in homogeneous solution; excellent enantioselectivities (96–98%) and good conversions (up to 98%) were obtained for the enone epoxide. Using these improved methods the scope of Julià-Colonna epoxidation has been extended to ketones with aliphatic substituents, symmetrical α,β-unsaturated ketones, dienes, and *bis*-dienes.[48,51]

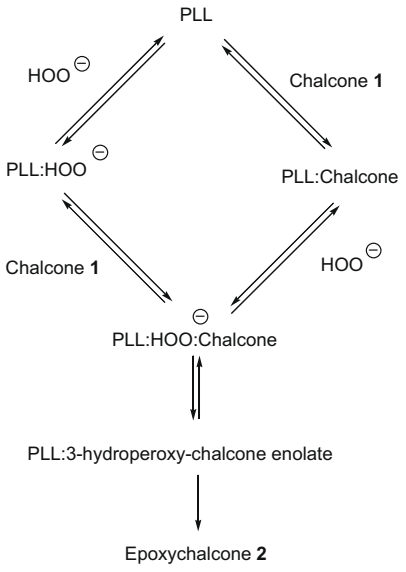
More recently, Gerlach and Geller reported the preparation of L-leucine NCA on a multi-100 g scale, which was polymerized using DAP in toluene. The poly-L-leucine was used directly for the epoxidation of a chalcone on a 100-g scale. Optimum conditions were found to entail the use of only a small amount of catalyst (0.35 mol %), together with a phase-transfer agent (such as tetrabutylammonium bromide) in hexane and aqueous sodium hydroxide (1.3 equiv) containing hydrogen peroxide (5 equiv) at 25 °C over 20 h. The reported yield of epoxychalcone **2** (95% ee) was 75% and the recovered catalyst could be reused successfully.[52] This methodology is particularly useful for large scale work since it allows the recycling of the insoluble catalyst, the minimal use of organic solvents, and a massive enhancement of the reaction rate with almost complete enantioselectivity.

4. Mechanistic considerations

The PEG-bound polyleucine (PLL) has the big advantage of total solubility in several organic solvents. This has led to a breakthrough in the understanding of the role of polyamino acids in the Julia-Colonna epoxidation reaction and shown that they behave as real synthetic enzymes. The direct investigation of the kinetics for the epoxidation of chalcone **1** was indeed possible under monophasic conditions using PEG-PLL, UHP, and BEMP, a strong base (2-*t*-butylimino-2-diethylamino-1,3-dimethyl-perhydro-1,3,2-diazaphosphorine, BEMP) for inducing the dissociation of hydrogen peroxide in THF as solvent.

The results indicated that PLL behaves as an enzyme-like catalyst, at relatively low concentrations of substrates, and shows saturation kinetics for both chalcone (K_M 110 mM) and HOO^- (K_M 30 mM).[53,54]

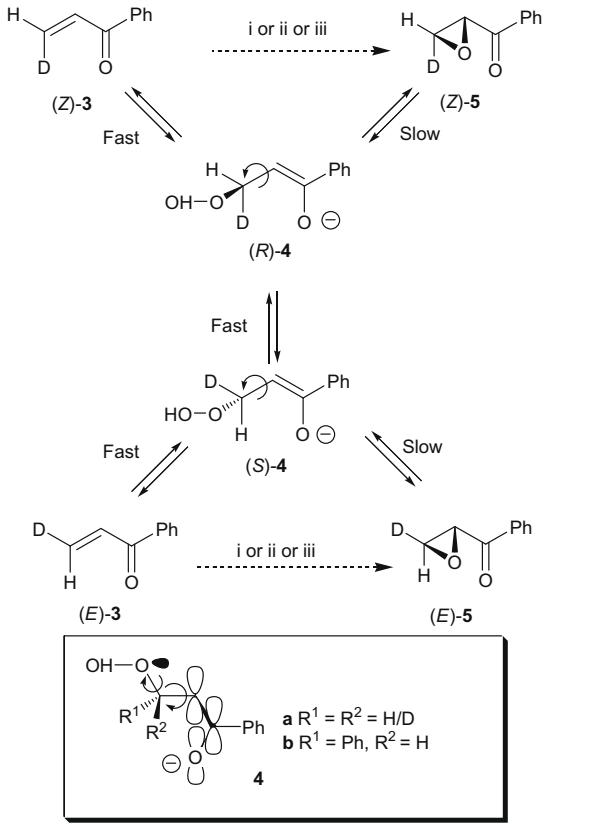
The observation of an induction period offers a rationalization for effects formerly attributed by us to substrate inhibition (Scheme 3).[55]



Scheme 3. Kinetics for chalcone oxidation by hydrogen peroxide anion catalyzed by poly-L-leucine (PLL).

The lower K_M for hydroperoxide anion is in agreement with its sequestration by polyleucine and there is no indication of any other significant intermediates. Hence the peroxy-enolate formed is transitory and irreversibly gives the epoxide as the reaction product when such enolate has achieved the geometry required for its formation. As suggested by Ferdinand for a two substrate enzyme,[56] the results can be explained on the basis of a steady state random bireactant mechanism, which implies pathways to the ternary complex and, very importantly, postulates that the pathway (HOO^- binding first) is the only one kinetically preferred whereas the other (chalcone binding first)[53,54] (Scheme 3) is negligible.

The Weitz-Scheffer epoxidation[57,58] of electron-deficient alkenes by alkaline hydrogen peroxide is the achiral progenitor of the Julià-Colonna epoxidation. It is rationalized as a Michael-type attack of hydroperoxide anion at the β-position to give a peroxy-enolate. This is followed by intramolecular nucleophilic displacement at the proximal oxygen, with ejection of the hydroxide group. The fast and reversible addition of hydroperoxide and the slow ring closure to afford the epoxide justify the formation of the *trans*-chalcone epoxide under Weitz-Scheffer and Julià-Colonna conditions, from both the *cis*- and the *trans*-chalcone. By monitoring the epoxidation of (*Z*)-3-deuterated-phenylprop-2-enone by ^1H NMR it was found that in both the Weitz-Scheffer and Julià-Colonna reaction isomerization of the alkene occurred rapidly and equimolecular mixtures of (*Z*) and (*E*) epoxide were formed[59] (Scheme 4).



Scheme 4. Mechanistic scheme for the Weitz-Scheffer and Julià-Colonna epoxidations.

Soluble PEG-polyleucine containing as few as five residues has catalytic activity (rate and enantioselectivity), which is comparable to that of insoluble polyleucine with 20+ residues.[60,61] Four leucine residues attached to aminoPEG catalyzed the epoxidation of chalcone with 42% ee, while five residues gave 87% ee. With six residues the enantioselectivity was the same of that observed previously with longer catalysts.[60] Similar results were found by

Berkessel et al., using stepwise prepared catalysts.[61] The fact that the epoxidation activity is independent of the nature of the initiating amine, which resides at the C-terminus as a carboxamide, indicates that the catalytic site is located at the N-terminus. *N*-acetyl and more importantly the *N*,*N*-dimethyl oligopeptides are still catalytically active; therefore a significant role of the N-terminal amino group as hydrogen bond donor is very unlikely.[62,44] This behavior was confirmed by the use of oligopeptides containing various combinations of L- and D-amino acids. Also in these cases the stereochemistry of the epoxide was determined by the stereochemistry of the N-terminal residues.

Leucine has close to the highest α-helix propensity of all natural amino acids and the predominance of this conformation in solution for the soluble catalysts has been shown using CD.[60] The four N-terminal N–H groups of an α-helical polypeptide chain are not engaged in intrachain hydrogen bonds, thus providing an ideal set of acceptors for hydrogen bonding (an oxyanion hole) to enones **1** and **3** and, more importantly, peroxyenolates **4**.

In this respect the model proposed by us (Fig. 1) has the advantage that only the amidic N–H groups act as hydrogen donors and these are much more efficient donors than an amino group such as NH-1. There has been much interest in the last few years in the chiral amplification for practical synthetic reasons and for its possible role in the origin of life. The Julià-Colonna epoxidation was the first example of a chiral amplification system, which does not involve a metal. When chalcone **1** was epoxidized using a catalyst prepared from a mixture of D- and L-NCA (20% ee) the resulting epoxide 2 had an enantiomeric excess higher than 73.5% and almost full enantioselectivity was only observed with a catalyst obtained by monomers with 43% ee.

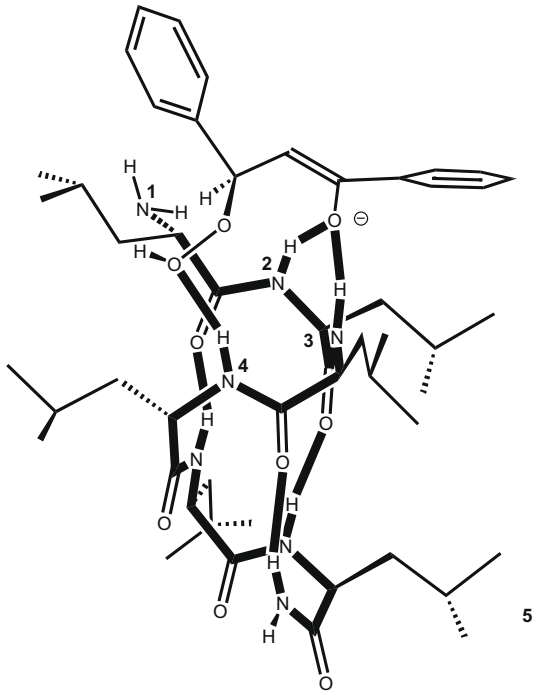


Figure 1. Hexa(L-leucine) carboxamide, 3-hydroperoxy-chalcone enolate complex **5**.

The fact that a low molecular weight polymer of five or more amino acids, not necessarily enantiomerically pure, can act as a synthetic enzyme and afford a product with much higher enantiomeric excess could be an indication that this kind of chiral amplification might have played a role in similar systems under prebiotic conditions.

Other enzyme-like catalysts (also known as 'synzymes' or 'enzyme mimetics'), each comprising of a small number of amino acid residues, have featured in the literature recently. For instance, tetrapeptides, designed by Miller et al.,[63] effect enantioselective acylation of some chiral cyclohexanol derivatives, mimicking the well-known transformations of lipases and other hydrolases in solvents of low water activity. The selectivity of hydroxynitrile lyases (e.g., from *Prunus* and *Hevea* species) is replicated to a large extent by some selected cyclic dipeptides, as discussed for [D-His-D-Phe].[23]

Leucine and many other α-amino acids were likely to have been present in the prebiotic soup, in agreement with Miller-Urey experiments. The importance of leucine is stressed by the fact that leucine is the most common amino acid in contemporary proteins (9.1%) and alanine is the second most (7.8%).[38]

Prosite enolase is comparatively leucine rich and, although there is no evidence that it is related to phosphoenolpyruvate, it is tempting to evoke a similar behavior of these two enzymes. The Michael addition of water catalyzed by enolase is a key step in glycolysis, this ubiquitous enzyme being the most highly conserved of all glycolytic enzymes.

5. Conclusions

Although chalcone epoxide is not a reliable candidate in the prebiotic soup, it is a product of the Michael addition reaction of hydrogen peroxide. The aldol reaction is certainly not only a key reaction in organic synthesis, since it forms C–C bonds, but also has a remarkable biological and prebiological significance.[24] Polyleucines of various lengths act as enantioselective catalysts in aldol condensations between cyclohexanone and a series of aromatic aldehydes, as discussed above, which may be of prebiotic significance.

It is worthwhile mentioning a recent paper dealing with the origins of the stereoselection of the dipeptide-catalyzed intermolecular reaction between cyclohexanone and benzaldehyde by means of a hybrid density functional system.[64] The calculations again demonstrated that the main source of stereoselectivity is the interaction of the N-terminal amino acid side chain of (*S*)-ala(*S*)-ala with the cyclohexane ring, with possible implications for the evolution of homochirality.

In the transformation from racemic to chiral biology, the spontaneous segregation of enantiomers from a racemic mixture in monolayer aggregates could have played an important role.[65] Interestingly this hypothesis is supported by the generation of oligopeptides with homochiral sequences in the polymerization of racemic mixtures of monomers segregated into two dimensional enantiomorphous domains at the water–air interface.[66]

Darwin evoked a 'warm little pond'.[67] as the first cradle. In this still poorly defined environment, processes had to take place which were capable of maintaining the self-sustaining systems that still replicate and evolve chemical information, as life is defined in its broadest sense.

A top down logics suggest that in the pond at one point nucleic acid-based genotypes had to cooperate and coevolve with protein-based phenotypes. The processes described here show that amino acids and oligopeptides are endowed with the pristine property of inducing and expanding selectivity.

References

1. Lincoln, T. A.; Joyce, G. F. *Science* **2009**, *232*, 1229.
2. Mansy, S. S.; Schrum, J. P.; Krishnamurthy, M.; Tobe, S.; Treco, D. A.; Szostak, J. W. *Nature* **2008**, *454*, 122.
3. Saladino, R.; Crestini, C.; Ciciriello, F.; Costanzo, G.; Di Mauro, E. *Chem. Biodivers.* **2007**, *4*, 694.

4. Saladino, R.; Neri, V.; Crestini, C.; Costanzo, G.; Graciotti, M.; Di Mauro, E. *J. Am. Chem. Soc.* **2008**, *130*, 15512.
5. Costanzo, G.; Saladino, R.; Crestini, C.; Ciciriello, F.; Di Mauro, E. *J. Biol. Chem.* **2007**, *282*, 16729.
6. Powner, M. W.; Gerland, B.; Sutherland, J. D. *Nature* **2009**, *459*, 239.
7. Miller, S. L. *Science* **1953**, *117*, 528.
8. Aubrey, A. D.; Cleaves, H. J.; Bada, J. L. *Origins Life Evol. Biosphere* **2009**, *39*, 91.
9. Johnson, A. P.; Cleaves, H. J.; Dworkin, J. P.; Glavin, D. P.; Lazcano, A.; Bada, J. L. *Science* **2008**, *322*, 404.
10. Cleaves, H. J.; Aubrey, A. D.; Bada, J. L. *Origins Life Evol. Biosphere* **2009**, *39*, 109.
11. Bonner, W. A. *Origins Life Evol. Biosphere* **1996**, *21*, 59.
12. Leman, L. J.; Orgel, L. E.; Ghadiri, M. R. *Science* **2004**, *306*, 283.
13. Leman, L. J.; Orgel, L. E.; Ghadiri, M. R. *J. Am. Chem. Soc.* **2006**, *128*, 20.
14. Huang, Z. Z.; Leman, L. J.; Ghadiri, M. R. *Angew. Chem., Int. Ed.* **2008**, *47*, 1758.
15. Noz, W.; Tanaka, F.; Barbas, C. F., III *Acc. Chem. Res.* **2004**, *37*, 580.
16. Blackmond, D. G.; Klussmann, M.; Iwamura, H.; Mathew, S. P.; Wells, D. H., Jr.; Pandya, U.; Armstrong, A. *Nature* **2006**, *441*, 621.
17. Hayashi, Y.; Matsuzawa, M.; Yamaguchi, J.; Yonehara, S.; Matsumoto, Y.; Shoji, M.; Hashizume, D.; Koshino, H. *Angew. Chem., Int. Ed.* **2006**, *45*, 4593.
18. Breslow, R.; Levine, M. S. *Proc. Natl. Acad. Sci. U.S.A.* **2006**, *103*, 12979.
19. Blackmond, D. G. *Chem. Eur. J.* **2007**, *13*, 3290.
20. Naithani, V. K.; Dhar, M. M. *Biochem. Biophys. Res. Commun.* **1967**, *29*, 368.
21. Moser, R.; Thomas, R. S.; Gutte, B. *FEBS Lett.* **1983**, *57*, 247.
22. Shen, C.; Lazcano, A.; Orò, J. *J. Mol. Evol.* **1991**, *31*, 445.
23. Kogut, E. F.; Thoen, J. C.; Lipton, M. A. *J. Org. Chem.* **1998**, *63*, 4604.
24. Mahrwald, R. In *Modern Aldol Reactions* 2 Volume Set; Mahrwald, R., Eds.; Wiley-VCH, Weinheim, 2004.
25. Hayashi, Y. *Angew. Chem., Int. Ed.* **2006**, *45*, 8103. and references therein.
26. Klussman, M.; Iwamura, H.; Matthew, S.; Wells, D.; Pandya, U.; Amstrong, A.; Blackmond, D. *Nature* **2006**, *441*, 621.
27. Mukaiyama, T. *Angew. Chem., Int. Ed.* **2004**, *43*, 5590.
28. Enders, D.; Grondhal, C. *Angew. Chem., Int. Ed.* **2005**, *117*, 1235.
29. List, B. *Acc. Chem. Res.* **2004**, *37*, 548.
30. Berkessel, A.; Groger, H. *Asymmetric Organocatalysis*; Wiley: Weinheim, 2005.
31. Klussman, M.; White, A. J. P.; Armstrong, A.; Blackmond, D. G. *Angew. Chem., Int. Ed.* **2006**, *45*, 7985.
32. Hayashi, Y.; Aratake, S.; Itoh, T.; Okano, T.; Sumiya, T.; Shoji, M. *Chem. Commun.* **2007**, 957.
33. Weber, A.; Pizzarello, S. *Proc. Natl. Acad. Sci. U.S.A.* **2006**, *103*, 1151.
34. Weber, A.; Pizzarello, S. *Proc. Natl. Acad. Sci. U.S.A.* **2006**, *103*, 12713.
35. Cordova, A.; Zou, W.; Dziedzic, P.; Ibrahem, I.; Reyes, E.; Zou, Y. *Chem. Eur.* **2006**, *12*, 5383.
36. Carrea, G.; Ottolina, G.; Lazcano, A.; Pironti, V.; Colonna, S. *Tetrahedron: Asymmetry* **2007**, *18*, 1265.
37. Miller, S.; Lazcano, A. In *Life Origin: The Beginning of Biological Evolution*; Schopf, J. W., Ed.; California University: Berkeley, 2002; p 78.
38. Kelly, D. R.; Meek, A.; Roberts, S. M. *Chem. Commun.* **2004**, 2021.
39. Dziedzic, P.; Zou, W.; Ibrahem, I.; Sundén, H.; Cordova, A. *Tetrahedron Lett.* **2006**, *47*, 6657.
40. Jacobsen, E. N.; Wu, M. H.. In *Comprensive Asymmetric Catalysis*; Jacobsen, E. N., Pfaltz, A., Yamamoto, H., Eds.; Springer: Heidelberg, 1999; Vol. 2, pp 649–677.
41. Shi, H. *Acc. Chem. Res.* **2004**, *37*, 497.
42. Kelly, D. R.; Roberts, S. M. *Biopolymers (Peptide Science)* **2006**, *84*, 74.
43. Julià, S.; Masana, J.; Vega, J. C. *Angew. Chem., Int. Ed.* **1980**, *19*, 929.
44. Julià, S.; Guixer, J.; Masana, J.; Rocas, J.; Colonna, S.; Annunziata, R.; Molinari, H. *J. Chem. Soc., Perkin Trans. 1* **1982**, 1317.
45. Allen, V.; Bergeron, S.; Griffiths, M. J.; Mukherjee, S.; Roberts, S. M.; Williamson, N. M.; Wu, L. E. *J. Chem. Soc. Perkin Trans. 1* **1998**, 3171.
46. (a) Itsuno, S.; Sakakura, M.; Ito, K. *J. Org. Chem.* **1990**, 6047; (b) Cappi, M. W.; Chen, W. P.; Flood, R. W.; Liao, Y. W.; Roberts, S. M.; Skidmore, J.; Smith, J. A.; Williamson, N. M. *Chem. Commun.* **1998**, 1159.
47. Geller, T.; Roberts, S. M. *J. Chem. Soc., Perkin Trans. 1* **1999**, 1397.
48. Porter, M. J.; Skidmore, J. *Chem. Commun.* **2000**, 1215.
49. Flood, R. W.; Geller, T. P.; Petty, S. A.; Roberts, S. M.; Skidmore, J.; Volk, M. *Org. Lett.* **2001**, *3*, 683.
50. Tsogoeva, S. B.; Woltinger, J.; Jost, C.; Reichert, D.; Kuhnle, A.; Krimmer, H. P.; Drauz, K. *Synlett* **2002**, 707.
51. Lasterra-Sanchez, M. E.; Roberts, S. M. *Curr. Org. Chem.* **1997**, *1*, 187.
52. Gerlach, A.; Geller, T. *Adv. Synth. Catal.* **2004**, 1247.
53. Carrea, G.; Colonna, S.; Meek, A. D.; Ottolina, G.; Roberts, S. M. *Chem. Commun.* **2004**, 1412.
54. Carrea, G.; Colonna, S.; Meek, A. D.; Ottolina, G.; Roberts, S. M. *Tetrahedron: Asymmetry* **2004**, *15*, 2945.
55. Mathew, S.; Gunathilagan, S.; Roberts, S. M.; Blackmond, D. G. *Org. Lett.* **2005**, *22*, 4847.
56. Ferdinand, W. *Biochem. J.* **1966**, *98*, 278.
57. Weitz, E.; Scheffer, A. *Ber.* **1921**, *54*, 2327.
58. Bunton, C. A.; Minkoff, G. J. *J. Chem. Soc.* **1949**, 665.
59. Kelly, D. R.; Caroff, E.; Flood, R. W.; Heal, W.; Roberts, S. M. *Chem. Commun.* **2004**, 2016.
60. Kelly, D. R.; Bui, T. T.; Caroff, E.; Drake, A. F.; Roberts, S. M. *Tetrahedron Lett.* **2004**, *45*, 3885.
61. Berkessel, A.; Gasch, N.; Glaubitz, K.; Koch, C. *Org. Lett.* **2001**, *3*, 3839.
62. (a) Hol, W. G. *J. Prog. Biophys. Mol. Biol.* **1985**, *45*, 149; (b) Bentley, P. A.; Flood, R. W.; Roberts, S. M.; Skidmore, J.; Smith, C. B.; Smith, J. A. *Chem. Commun.* **2001**, 1616; (c) Banfi, S.; Colonna, S.; Molinari, H.; Julia, S.; Guixer, J. *Tetrahedron* **1984**, *40*, 5207.
63. Jarvo, E. R.; Miller, S. J. *Tetrahedron* **2002**, *58*, 2481.
64. Hammar, P.; Cordova, A.; Himo, F. *Tetrahedron: Asymmetry* **2008**, *19*, 1617.
65. Barocci, S.; Ceccacci, F.; Cruciani, O.; Mancini, G.; Sorrenti, A. *Synlett.* **2009**, 1023, and references therein.
66. Weissbuch, I.; Bolbach, G.; Zepik, H.; Shavit, E.; Tang, M.; Frey, J.; Jensen, T. R.; Kjaer, K.; Liserowitz, L.; Lahav, M. *J. Am. Chem. Soc.* **2002**, *124*, 9093.
67. Darwin, F. In *The life and letters of Charles Darwin*, London, Murray, J., Ed.; London, **1888**, 3, pp 18.