65 Marler, P.R., and Tamura, M., Culturally transmitted patterns of vocal behavior in sparrows. Science *146* (1964) 1483–1486.
66 Avise, J.C., Patton, J.C., and Aquadro, C.F., Evolutionary genetics of birds. II. Conservative protein evolution in North American sparrows and relatives. Syst. Zool. *29* (1980) 123–130.
67 Avise, J.C., Aquadro, C.F., and Patton, J.C., Evolutionary genetics of birds, V. Genetic distances within Mimidae (mimic thrushes) and Vireonidae (Vireos). Biochem. Genet. *20* (1982) 95–104.
68 Wilson, A.C., Sarich, V.M., and Maxson, L.R., The importance of gene rearrangement in evolution: evidence from studies on rates of chromosomal, protein and anatomical evolution. Proc. natl Acad. Sci. USA *71* (1974) 3028–3030.
69 Nobs, M.A., Experimental studies on species relationships in Ceanothus. Carnegie Inst. Wash. publ. 623, Washington, D.C. 1963.
70 Stebbins, G.L., Plant speciation; in: Mechanisms of Speciation, pp. 21–39. Ed. C. Barigozzi. Alan R. Liss, New York 1982.
71 Cronquist, A., An Integrated System of Classification of Flowering Plants. Columbia, New York 1981.
72 Mayr, E., Systematics and the Origin of Species. Columbia, New York 1942.

0014-4754/83/080823-12$1.50 + 0.20/0
©Birkhäuser Verlag Basel, 1983

Molecular selforganization and genesis of life

by H. Kuhn and J. Waser

Max-Planck-Institut für biophysikalische Chemie, D-3400 Göttingen-Nikolausberg (Federal Republic of Germany), and 6120 Terryhill Drive, La Jolla (California 92037, USA), formerly California Institute of Technology, Pasadena (USA)

The origin of life has presented challenging problems for modern biologists, because it is difficult to account for the development of even the simplest of living organisms. Beginning with Oparin[1], many modern authors[2–5] have attacked this problem. Present-day mechanisms of replication and translation of genetic information must have developed through many stages, through mutation and culling out by natural selection. A fundamental difficulty is that living systems operate by means of enzymes, the manufacture of which requires many enzymes in the first place.

It is our contention that a fruitful approach to this problem is to consider a model consisting of many small but physically and chemically plausible steps. It is easy enough to claim (erroneously) that the origin of life could never be explained on the basis of physical laws simply by considering some large step and showing that its probability is ridiculously small. However, it turns out that the probabilities for the steps we consider are of an appropriate order of magnitude to assure near certainty for their occurrence under model conditions[6–10].

Model requirements

Living organisms consist of macromolecules that act together like the components of a machine. Some of these molecules are capable of replication and moreover contain the blueprints of the machine, permitting its reproduction. The emergence of such systems in their most primitive form requires the presence of a) a structurally diversified environment as that which existed, for example, on prebiotic earth in form of porous rock formations; b) a suspension of energy-rich nucleotide-like compounds presumed to have existed on prebiotic earth; and c) a regime of periodic temperature changes as caused, for example, by the rhythm of light and shadow generated by the daily motion of the sun and by local shadow-casting objects. Inside suitable pores, such conditions favor the creation and later drive the replication of short polymer strands, with (+) strands acting as templates for (−) strands and vice versa. Pores above a certain size cannot be colonized by such strands, even though sufficient monomers are present, because strands that are too short diffuse too quickly from the minute region in which the required regime of critical temperature changes exists. In the model, short strands are lengthened by repeated doubling, thereby coloninzing larger pores, but there is a limit to the number of monomers a replicable strand may contain because of the unavoidability of copying errors. That is, the probability that replicate strands be error free must be large enough to assure their survival.

An error filter

In the model, the resulting impasse is broken by a mechanism in which the strands convolute into the conformation of a hairpin (fig. 1) with legs twisted into a double helix. Such hairpin strands would be capable of interlocking into picket-fence like aggregates (fig. 2), stabilized perhaps by bivalent cations, such as Mg^{+2} or Ca^{2+} ions, that form links between negatively charged phosphatidyl groups on the outside of neighboring hairpin strands. Not only is diffusion of such aggregates slowed by their size, but it is of the greatest importance that practically all erroneous copies are rejected during aggregation because of the precision required for interlocking. Aggregation therefore serves the function of an error filter.

A great selective premium is associated with the evolution of machinery that facilitates the aggregation of interlocking subunits B_1, B_2, B_3, ... A model of such machinery, which we call the assembler, is that of a convoluted strand A with an unfolded strand C protruding from one of its ends. The function of C is that of a collector strand for the subunits B_1, B_2, B_3, ... that are guided by this strand to the growth region of the aggregate, being bonded to strand C at times and not bonded to it at other times, spatial diffusion of the subunits being converted in this way into diffusion along the strand. An important function of A is postulated to be that of a nucleation site, facilitating the onset of aggregation and acting as endpost for the picket-fence like aggregate.

A detailed molecular model of the assembler

A detailed model with GC RNA strands in the conformation of a left-handed double helix analogous to the 'Z' conformation established by Rich, Arnott, and by Dickerson and their co-workers[11] for GC DNA strands, exhibits an astonishingly good fit between neighboring hairpin subunits (fig. 3). Equally excellent is the steric fit between 'anticodon' loops that form the hairpin bends and 'codons' along the collector strand. The bases in these complementary triplets are stacked, providing stabilization, and the 3′5′ directions of the strands connected by complementary base triplets are opposite. The steric fit between complementary bases along the 180° hairpin bend and the collector strand appears to be ruined if more than 3 nucleotide pairs are involved in the attachment of subunits to collector strand, while a 180° hairpin bend requires at least 3 nucleotides. Finally there is an excellent fit between the hairpin strand of opposite orientation that functions as nucleation site A and the first subunit B_1. This fit requires, however, that the 3′5′ direction of the 2 strands is as indicated in figure 2. A molecular model based on a right-handed double helix (the 'A' conformation) is feasible also, but does not permit such close fits in the aggregate as can be achieved with a Z conformation.

It is known that the bonding energies of base pairing by hydrogen bonds are insufficient at room temperature to establish stable bonding between 2 triplets of complementary bases. The model would therefore permit the required one-dimensional diffusion of subunit pickets along the collector strand. There is, of course, little constraint on orientation, and any new picket that may be on its way towards being incorporated will point in more or less random directions during its travels to the growing aggregate. Even after having arrived at the aggregate it may still become detached, because of the weakness of its bonding to the collector strand. However, once the new hairpin molecule assumes an appropriate orientation by chance, it is now in a favorable location for joining up,

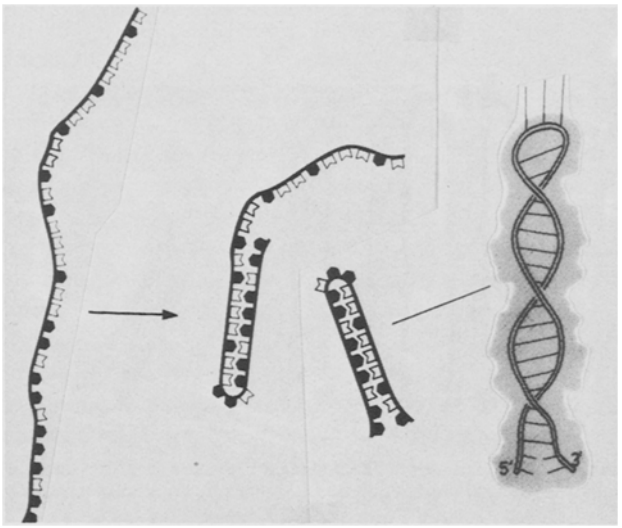

Figure 1. Open strand and nucleotide sequence that permits a hairpin conformation. Legs of hairpin twisted into double helix. The outlines are as expected for the van der Waals contacts based on a molecular model (cf. fig. 3).

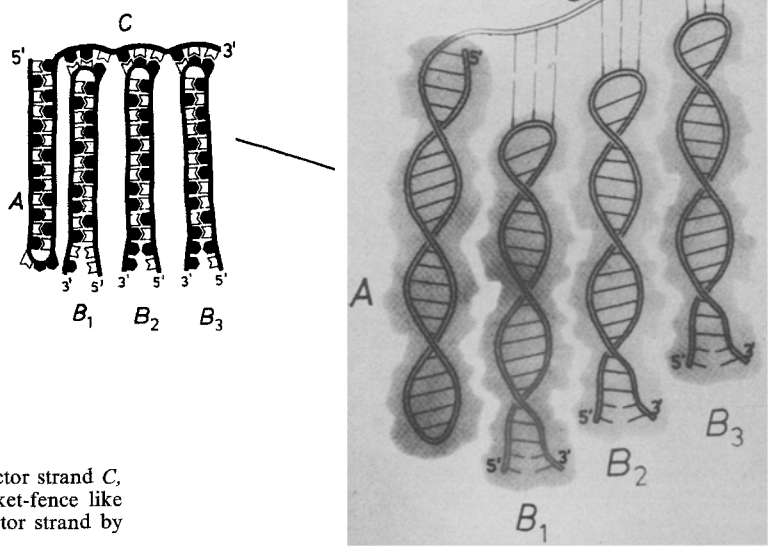

Figure 2. Schematic view of nucleation unit A, collector strand C, and subunits B_1, B_2, B_3, capable of forming a picket-fence like aggregate. Eventual attachment of subunits to collector strand by base triplets.

provided it is capable of a close enough fit to the picket preceding it along the strand. Studies with molecular models show, for example, that the lateral contacts between hairpin strands consisting of some 20 nucleotide pairs are such as to permit the interlinkage of about 10 phosphatidyl groups by Me^{2+} ions. This, together with the linkage to the collector strand by the 3 required complementary base pairs is sufficient for the integration of the new picket unit into a quite rigid aggregate. A strand with an imperfect hairpin conformation would, however, not be capable of forming an adequate number of $O-Me^{2+}-O$ linkages and would be rejected. And of course, even a perfect hairpin strand could be incorporated into the aggregate only if it carried the appropriate bases along its 'anticodon' loop, complementary to the bases of the corresponding 'codon', and not otherwise.

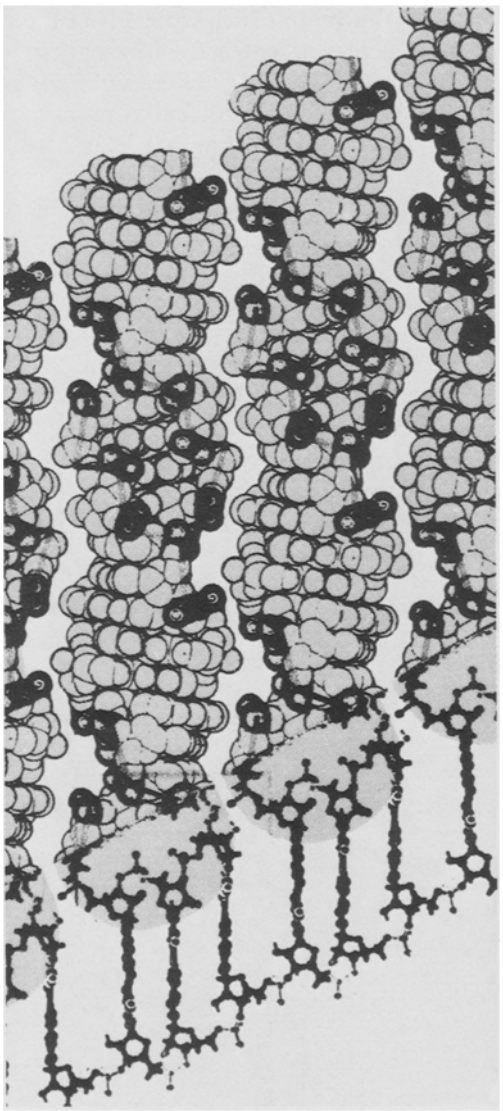

Figure 3. Details of the excellent fit between the different hairpin strands in picket-fence like aggregate and between the base triplets at the hairpin bends and complementary triplets along the collector strand.

Evolution of catalytic properties

There exists selective pressure towards the invasion of pores that contain sufficient monomers, but which are too large to curb strand diffusion effectively. It is proposed that such pores can be colonized with the aid of polypeptides formed, by catalytic action of the aggregates, from amino acids presumed to be present on prebiotic earth. Such polypeptides could form impediments in pore channels that curb diffusion; later they could coalesce into confining 'cellular' envelopes that keep strands together as aggregates disassemble into strands which replicate and again aggregate. A strong evolutionary gradient would exist in this manner.

In our model the aggregates described develop catalytic properties in the following way. The open ends of the hairpin strands possess or develop affinity towards activated amino acids that are thereby brought into favorable proximity of each other and enabled to form polypeptide bonds; that is, the hairpin strands become adapters of amino acids. Open ends of the hairpin strands would be of help in strand replication

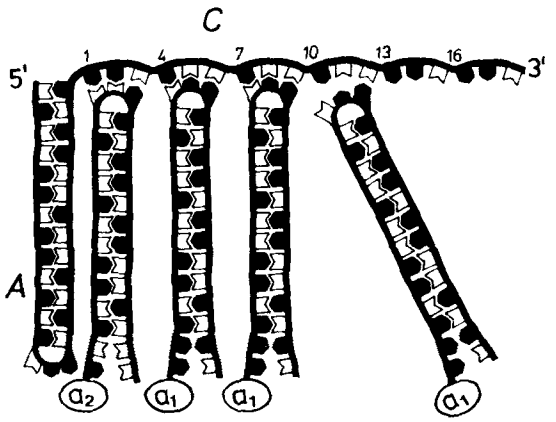

Figure 4. Picket-fence like aggregate. Amino acids a_1 and a_2 attached to the 3′ end of (+) and (−) hairpin strand respectively.

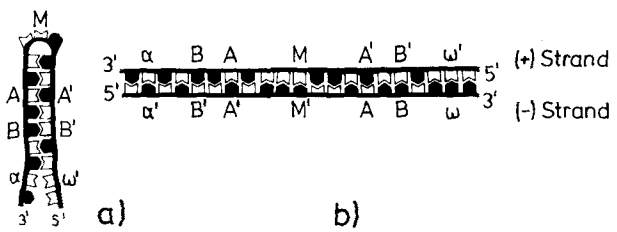

Figure 5. Replication of hairpin strands. *a* Primed and unprimed letters represent complementary bases. A requirement for the hairpin conformation is that any two juxtaposed base pairs along the legs, such as A and A′ or B and B′, are complementary. One or more bases at the ends of the legs, symbolized by α and ω' need not be paired, and the base M at the midposition of the hairpin loop has no juxtaposed partner. *b* Replication, in which the hairpin strand depicted in (a) serves as template, requires complementarity of bases as indicated and leads to the replicate (−) strand shown. Comparison reveals that the bases A, B, B′, A′ occur in the same sequence along the two strands, so that (+) and (−) strands are identical, except for M, α, and ω. Note that the 3′5′ directions of template and replicate must be opposite for this near-identity to come about.

as well, because replication could automatically begin at an end of a strand only and selective advantages would be gained in this way.

An attractive feature of the hairpin conformation is that (+) and (−) strands are identical except for the nucleotide in the middle (the midposition of the anticodon loop) and the nucleotides at the open ends (fig. 4). This is because of what is known as the 3′5′ direction of the replicate obtained in the template-assisted polymerization is opposite to the 3′5′ direction of the template strand (fig. 5). Hence the (+) and (−) strands would be capable of serving as adapters for 2 different kinds of amino acids - a feature that permits great economy in the usage of strand material and in the assemblage of aggregates. To fix ideas, let us assume that the midpositions as well as the ends of a (+) strand are occupied by C bases; there must then be G bases at the midpositions and at the ends of a (−) strand. The 2 kinds of strand ends could then have different affinities for 2 kinds of activated amino acids. Amino acids bonded to nucleotides could have existed prebiotically, for example, a_2 combined with C and a_1 with G. In this way (+) strands would serve automatically as adapters C_{a_2} for a_2 and (−) strands as adapters G_{a_1} for a_1 (fig. 4).

These requirements impose some restrictions on the nucleotide sequence along the collector strand. Assuming that the sequence (read in the 3′5′ direction) along the loop of the (+) strand is CCG (as in strand B_1 in fig. 4), then the sequence along the loop of the (−) strand is CGG (as e.g. in strand B_2 in fig. 4). At the positions 1, 4, 7, ... (fig. 4) of the collector strand (corresponding to the first position on the triplet of the hairpin loop) there must therefore be base G and at the positions 3, 6, 9, ... the base C. The mid position can randomly be occupied by G or C. In the 1st case a (+) hairpin strand would be attached to the collector strand, in the 2nd case a (−) strand. The bonding energies involved in base pairing are known to be insufficient at room temperature to establish stable bonding between two triplets of complementary bases G and C. Therefore a hairpin will become firmly attached to strand C only if it reaches the region of growth of the aggregate, if its shape allows for a close enough fit to be stably incorporated, and if its triplet is complementary to the corresponding triplet on filament C.

The charging of adapters with amino acids

The molecular model suggests the following mechanism for charging adapter molecules with amino acids. The amino acids become activated by becoming linked to nucleotides such as a_1 to C and a_2 to G. It has, for example been shown that adenosine (ado-) 5′-phosphoimidazolide forms aminocyl adenylates[12]. A specific linkage to the appropriate (+) and (−) adapter molecules is then achieved by intercalation and complementary base pairing[8] (fig. 6), a linkage that places the amino acids in a position favorable for reacting with the 2′-OH group of the ribose of the 3′-terminal nucleotide of the adapter molecule, the linkage that corresponds to that found in today's charged tRNA. (The model permits reaction with the 2′- as well as the 3′-OH groups of this ribose.)

Another aspect of the proposed mechanism for the charging of adapters concerns the activation of specific amino acids, with specific nucleotides. One possibility arises from the experimental observation[13] that under certain conditions the elutions by a dilute aqueous salt solution, of adsorbed glycine and the G-nucleotide GMP (guanosine-mono-phosphate) occur at approximately equal speeds, while alanine and the C-nucleotide CMP (cytidine-mono-phosphate) are eluted at another, significantly different, speed. This suggests that GMP-gly and CMP-ala might have been formed in the very pores in which the crucial action takes place, these pores being in the neighborhood of reservoirs in which the component amino acids and nucleotides are adsorbed in a suitable fashion.

Appearance of a replicase and stabilization of a genetic code

Eventually, by means of random copy errors, a nucleotide sequence will arise along the collector strand that corresponds to a polypeptide that exhibits enzymatic properties, acting as a primitive 'replicase'. Such a 'replicase' would have to decrease the frequen-

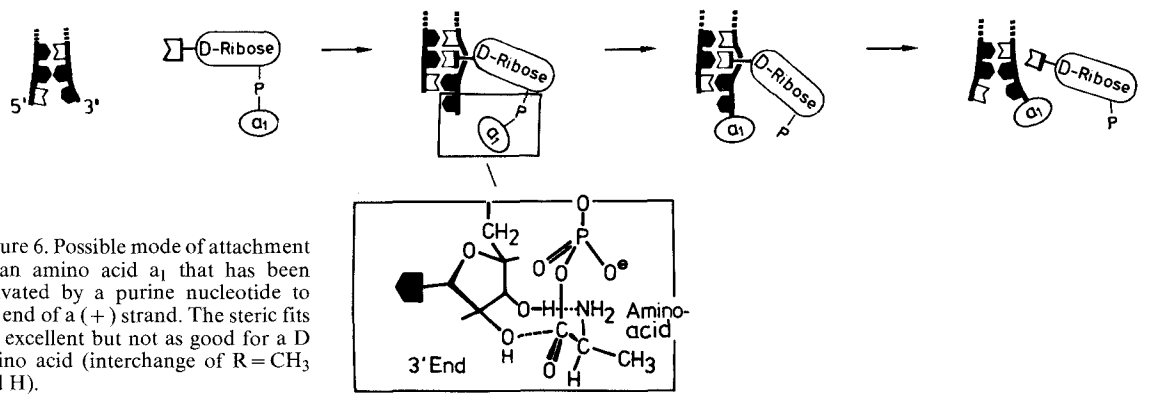

Figure 6. Possible mode of attachment of an amino acid a_1 that has been activated by a purine nucleotide to the end of a (+) strand. The steric fits are excellent but not as good for a D amino acid (interchange of R = CH_3 and H).

cy of replication errors sufficiently so that the information coding for it would not be lost during the number of generations required to fix it by selection. It can be shown, for example, that if the survival rate is increased by just 10% by the advent of a replicase consisting of 10 amino acids, a reduction of the replication error rate per base from 1/100 to 1/300 is sufficient for fixation by selection[9].

Such a replicase may, for example, be a suitable short polypeptide that fits into a notch of the double helix which exists during strand replication, in the region where the new strand is formed, slipping along in the notch to remain in the region of replication as the new strand is lengthened. It has, for example, been shown[14] that a section of a polypeptide of only some 10 amino acids can form a 2-stranded antiparallel β sheet that fits into the groove of a double helix.

The presence of a 'replicase' stabilizes the doublehelix conformation during replication and improves the contact between the 2 strands in that region. Replication is speeded up and there are fewer errors in base pairing during replication. The primitive 'enzyme' induces the transformation of the assembler into a code translator. The stabilization of machinery for reading and translating a code represents a major breakthrough, making possible the acquisition of further enzymes, and conferring stability upon the code.

The similarity between the functioning of the aggregates described here and the operation of present-day genetic machinery is striking, a feature of the model that is most encouraging if not altogether surprising. According to the model, the collector strand C would be the precursor of messenger RNA, the subunits B_i those of t-RNA's and the 'cellular' envelope that of the cell membrane. As is the case in today's biosystems, codons along C are read in the 5′3′ direction while for attached adapters B_i the strand direction is opposite. The nucleation unit A could be the precursor of the messenger strand that recognizes the start of the genetic message. In many details, including the hairpin conformation of adapter molecules and the use of the middle position of the triplet for ancestral coding, the apparatus discussed in the foregoing is identical with one described several years ago[5].

Some of the model assumptions have been supported by recent experimental results. A basic model aspect, the proposition that the transfer RNA's of all amino acids have a common ancestor, is well supported by comparative studies by Eigen and Winkler[15] of tRNA sequences indicating a common ancestral sequence, which consisted predominantly of G and C nucleotides, supporting the hypothesis that G and C were first used. The sequence is very similar to the sequence obtained by Kilpatrick and Walker[16] in an archebacterial tRNA, which is a very old organism in the phylogenetic sense. If a strand with such a sequence is assumed to be in a hairpin conformation we find purine-pyrimidine base pairs in 85% of all pairs. This supports the view that the ancestral adapters were in a hairpin conformation and that only a few pyrimidines were changed later to purines or purines to pyrimidines.

The view that the collector strand is the primordial form of the carrier of genetic information, given in the reading frame GNC (N = G or C), is supported by a sequence analysis of the DNA of virus, procaryotes, and eucaryotes by Shepherd[17]. The clearly found periodic correlations indicate that originally there existed the reading frame PuNPy (Pu = purine, like G; Py = pyrimidine, like C; N = purine or pyrimidine). The difference in the deviations from the PuNPy reading frame in different organisms was found to be correlated with the time of their phylogenetic diversion, and from this the calculated time of last use of the PuNPy frame was found to be of the order of $3 \cdot 10^9$ years ago, a number which agrees well with fossile traces of early life. It can easily be imagined that G and C were first used exclusively or predominantly and that later G was partially interchanged with another purine (A) because of its steric similarity, and C with another pyrimidine (U) leading to the PuNPy reading frame. However, a pyrimidine was rarely replaced by a purine and vice versa, leading from GGC and GCC to the general PuNPy reading frame (with N = purine or pyrimidine).

The precursor of the ribosome

It is postulated that at an early stage an additional hairpin molecule D became important in providing additional stability to the site at which a new amino acid is added to the growing polypeptide chain, at the bottom of the ribbon of hairpin molecules that form the code-translator (fig. 7a). This molecule D would thus act as a precursor of today's ribosome[18] and may indeed have played an important stabilizing role in the mechanism of proper aggregate formation right from the start, even before amino acids came into play.

Let the new peptide bond be formed between the amino acids a_i and a_{i+1}, carried respectively by the adapter hairpin B_i and B_{i+1}, amino acids designated by an index smaller than i having already been linked together into an as yet incomplete polypeptide chain. In the proposed model both the 3′ and the 5′ ends of D are formed by pyrimidine nucleotides, and D associates with the 2 adapter molecules B_{i+1} and B_i by being linked to the bases at their 3′ ends. (For the very first peptide link formed between a_1 and a_2, the role of what would be B_0 ($i-1=0$) is played by A. (Fig. 7b). The main direction of D is parallel to those of B_{i-1}, B_1, ..., and, viewed along this direction, B_{i-1}, B_i and D form an approximately equilateral triangle. In the molecular model hydrogen bonds to D stabilize a steric arrangement of a_i and a_{i+1} relative to each

other that favors the formation of a peptide bond between them, as indicated schematically in figure 7b. Once the peptide bond has been formed and the linkage of a_i to B_i has been broken, D also becomes detached and moves about until it has diffused into the region between the adapters and assists in the peptide bond formation between a_{i+1} and a_{i+2}, etc. The formation of the peptide bond according to this model is precisely the same as that in today's biosystems, linking the carboxy group of a_i with the amino group of a_{i+1}. This molecular model of the function of D may represent the early version of the remarkable mechanism of today's ribosome, in which adapter molecules are shifted from what is called the A site to the P site.

L versus D amino acids

An important aspect of the present model is that good steric fits and adequate hydrogen bond formation can be achieved only if all amino acids involved are of the L variety. Steric difficulties arise when D rather than L amino acids are used (interchange of H and R in the amino acids). This situation arises in the model mechanism described earlier, by which adapter molecules are charged with amino acids, and again for the mechanism just discussed that catalyzes the interlinkage of amino acids into polypeptides by the aid of the 'ribosome' D. These model aspects may well be at the root of the well-known fact that proteins consist exclusively of L-amino acids.

If all adapter hairpins were replaced by their mirror images, all L-amino acids would have to be replaced by D-amino acids, so that the model relates the exclusive L-amino acid make-up of proteins to the handedness or chirality of the adapter hairpins. How then did this chirality come about? It may well be related to the fact that a helical build-up of large units from small subunits is particularly efficient. This is because for each new subunit the geometry of insertion remains constant, analogous to the invariant relationship of each step in a circular stairway to the steps preceding it. Thus, in crystal growth, a helical buildup along a screw dislocation is often favored. The same principles apply to the way a strand can be built by joining monomers, as well as to the template replication of such a strand; in both situations a helical build-up is the most efficient one. For nucleic acids a helical build-up is additionally favored because it permits the stacking of nucleotide bases, affording special stability in this way.

Now the building components of a helix must all have the same chirality and the chirality of a nucleotide depends on that of its ribose component. Thus, given the requirement of helical growth, all nucleotides of a given RNA strand must contain either D-ribose (as is

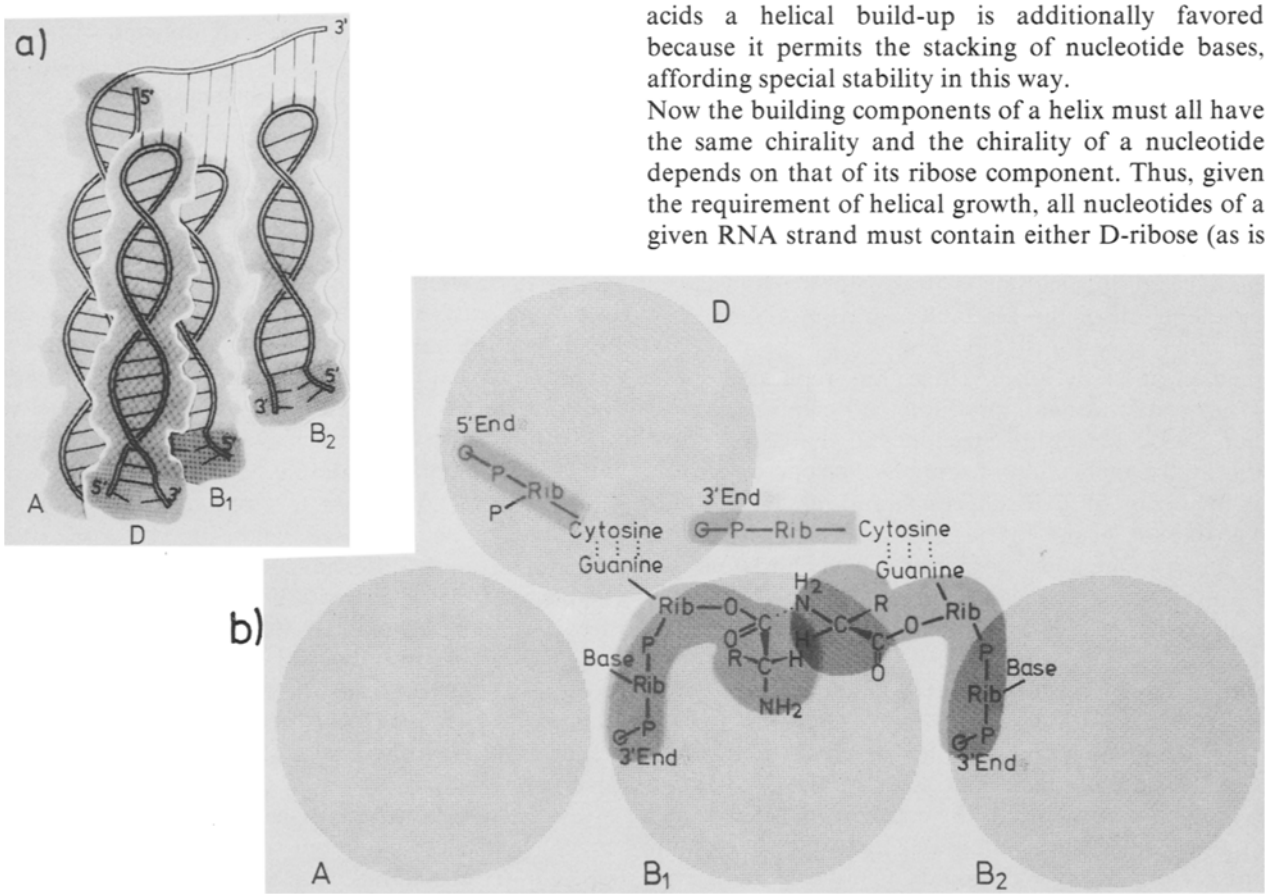

Figure 7. Possible role of primeval ribosome. *a* Hairpin molecule D as stabilizing factor in picket fence like aggregate. *b* Schematic view of peptide bond formation between amino acids linked to subunits B_1 and B_2. The C–O bond linking the amino acid to B_1 is broken and a peptide bond is formed between the carboxy C atom of this amino acid and the N atom of the amino acid linked to subunit B_2. The good steric fits are lost when a D rather than an L amino acid is used (interchange of R and H). The 'ribosome' D stabilizes an arrangement that favors peptide bond formation. The base of the 3′ end of the ribosome is paired with the base at the 3′ end of B_2, the base at the 5′ end of the ribosome is paired with the base at the 3′ end of B_1 (see ref. 18).

true of natural RNA), or all must contain L-ribose. Replication of this strand again requires nucleotides of the same chirality. Strands based on D-ribose thus evolved separately from strands based on L-ribose.

Once a replicable strand of either kind arose by chance at a given site, its descendants would consume all available common building elements in the neighborhood (phosphates and bases) so that no strand of the opposite chirality would arise in the close environment. A strand of the opposite chirality could arise elsewhere, of course, and there might be competition, at some time and location, between the descendants of the 2 types. However, sooner or later, one chirality would gain ascendancy over the other; the latest period during which one form would win out over the other is when enzymes would have evolved in the one that are capable of attacking the other.

According to these ideas, strands made of nucleotides containing D-ribose (or their equivalents) were formed originally or won out later on, and the chirality of hairpin adapters based on D-ribose then determined the chirality of the amino acids that became exclusive protein constituents. If the original strand had been based on L-ribose - the two possibilities have the same a priori probability - all proteins would now contain D-amino acids.

The genetic message is stored by DNA

Following this detailed description of the genesis and functioning of the code-translator, we shall describe some of the later steps in the evolution of early life more cursorily. Once a code-translator exists inside an envelope, all of the assemblers are quickly replaced by other code-translators. Because of the compartmentalization by an envelope, the 'replicase' that a given code-translator produces will catalyze replication inside the entire envelope, so that copy errors during the replication of some of the code-translators in the region of their collector strands no longer affect the viability of the entity, the 'organism' that consists of an envelope and its contents. Code-translators that furnish other enzymes can therefore evolve, and the complexity of the organisms can steadily increase. During the same period the genetic code becomes more complex; all 4 nucleotide kinds become part of it, as well as all 3 codon positions.

However, there is a limit to the complexity that can be reached on this evolutionary level. The reason is that only the (+) versions of the collector (or 'messenger') strands contain a meaningful 'genetic message', while the corresponding (−) strands only rarely do, if ever. Hence about half of the polypeptides synthesized by codetranslators are not useful; they crowd the interior of the envelope and waste precious building blocks (amino acids).

The problem can be overcome by a complete reorganization of the 'genetic apparatus' of the organism, in which one of the 2 roles that were hitherto played by RNA is taken over by DNA, while RNA keeps its function in the translation of a genetic message into a sequence of amino acids in a polypeptide. A mechanism, by which such a reorganization can take place by means of modifications of the original replicase, is described in detail elsewhere[9].

Interchange of genetic material

Another important limit to the complexity of an 'organism' arises at a later stage, because there is a limit to the amount of information such an organism can pass on to the next generation, as long as successive generations must meet the challenge of an ever-changing environment. This is because the recurrent adaptation of the system to environmental changes requires a minimum rate of copy errors, which in turn implies a maximum amount that can be passed by replication from generation to generation without being lost. That is, the probability W of a copy error in a given location must be small enough, so that the number of nucleotides along a strand endowed with a genetic message, N_{total}, must satisfy the order-of-magnitude relation $N_{total}W \simeq 1$ (Kuhn and Waser[9]). W must therefore decrease as the complexity of the 'organism', measured by N_{total}, increases. On the other hand, W must not be too small, so as to maintain a high enough frequency of 'mutations' to ensure adaptation to environmental changes.

It turns out that in order to maintain the information contained in a real protein, W must not be less than about 10^{-6}, which sets an upper limit of about 10^6 to the complexity measured by N_{total}. The only way to overcome the limitation imposed by this requirement appears to be through an interchange of genetic information (primitive 'sexuality'). Once this level of diversification has been achieved, the interchange of genetic material operates as a mechanism that is supportive of adaptation to environmental changes, and W can be decreased below the level of 10^{-6} without seriously affecting adaptation. In turn this makes possible an increase of N_{total} to values above 10^6, permitting an increase in complexity. Indeed, as N_{total} does increase beyond 10^6, W *must* show a corresponding decrease below 10^{-6} to maintain the relation $N_{total}W \simeq 1$ (by an increase in the precision of replication, by some error-correcting mechanism, or in other ways).

Eventually, W reaches a lower limit of about 10^{-8}, given by physical conditions such as by thermal noise, or by mutations induced by ever present background radiation. To overcome this limitation, it becomes necessary to move away from information storage on the molecular level. An 'artificial' means is required such as the use of written language, or now, of computer technology.

1 Oparin, A.I., The Origin of Life on Earth. Academic Press, New York 1957.
2 Katchalsky, A., Biological flow structures and their relation to chemicodiffusional coupling. Neurosci. Res. Prog. Bull. *9* (1972) 397–413.
3 Prigogine, I., Nicolis, G., and Babloyantz, A., Thermodynamics of evolution. Physics Today *25* (1972) Nov., pp. 23–28; Dec., pp. 38–44.
4 Eigen, M., Self-organization of matter and the evolution of biological macromolecules. Naturwissenschaften *58* (1971) 465–523; Eigen, M., and Schuster, P., The hypercycle – a principle of natural self-organization. Naturwissenschaften *65* (1978) 341–369.
5 Crick, F.H.C., Brenner, S., Klug, A., and Pieczenik, G., A speculation on the origin of protein synthesis. Origins of Life *7* (1976) 389–397.
6 Kuhn, H., Self-organization of molecular systems and evolution of the genetic apparatus. Angew. Chem., Int. Edn *11* (1972) 798–820.
7 Kuhn, H., Model consideration for the origin of life. Environmental structure as stimulus for the evolution of chemicals systems. Naturwissenschaften *63* (1976) 68–80; Evolution biologischer Information. Ber. Bunsenges. phys. Chem. *80* (1976) 1209–1223.
8 Kuhn, H., and Waser, J., Molecular self-organization and the origin of life. Angew. Chem., Int. Edn *20* (1981) 500–520.
9 Kuhn, H., and Waser, J., Self-organization of matter and early evolution of life; in: Biophysics, pp. 830–874. Eds W. Hoppe, W. Lohmann, H. Markl and H. Ziegler. Springer, New York 1982.
10 Kuhn, H., and Waser, J., Evolution of early mechanisms of translation of genetic information into polypeptides. Nature *298* (1982) 585–586.
11 Wang, A.H.-J., Quigley, G.J., Kolpak, F.J., Crawford, J.L., van Boom, J.H., van der Marel, G., and Rich, A., Molecular structure of a left-handed double helical DNA fragment at atomic resolution. Nature *282* (1979) 680–686; Drew, H., Takano, T., Takano, S., Itakuro, K., and Dickerson, R.E., High-salt d(CpGpCpG), a left-handed Z'DNA double helix. Nature *286* (1980) 567–572; Arnott, S., Chadrasekaran, R., Birdsall, D.L., Leslie, A.G.W., and Ratliff, R.L., Left-handed DNA helices. Nature *283* (1980) 743–745.
12 Lohrmann, R., and Orgel, L.E., Prebiotic activation processes, Nature *244* (1973) 418–420.
13 Lehmann, U., private communication.
14 Anderson, W.F., Ohlendorf, D.H., Takeda, Y., and Mattheus, B.W., Structure of the cro repressor from bacteriophage λ and its interaction with DNA. Nature *290* (1981) 754–758.
15 Eigen, M., and Winkler, R., Transfer-RNA: The early adaptor. Naturwissenschaften *68* (1981) 217–228; Transfer-RNA, an early gene? Naturwissenschaften *68* (1981) 282–292.
16 Kilpatrick, M.W., and Walker, R.T., The nucleotide sequence of the $tRNA_M^{Met}$ from the archaebacterium *Thermoplasma acidophilum*. Nucl. Acids Res. *9* (1981) 4387–4390.
17 Shepherd, J.C.W., Periodic correlations in DNA sequences and evidence suggesting their evolutionary origin in a commaless genetic code, J. molec. Evol. *17* (1981) 94–102; Method to determine the reading frame of a protein from the purine/pyrimidine genome sequence and its possible evolutionary justification. Proc. natl Acad. Sci. USA *78* (1981) 1596–1600; Shepherd, J.C.W., private communication.
18 Kuhn, H., Leben: Denkmodelle für die Entstehung lebender Systeme. Enzyklopädie Naturwissenschaft und Technik, Jahresband 1982. Verlag Moderne Industrie, Landsberg.

0014-4754/83/080834-08$1.50 + 0.20/0
©Birkhäuser Verlag Basel, 1983

Conclusion

by C. Petit*

Laboratoire de Génétique des Populations, Université Paris VII, 2, Place Jussieu, F-75221 Paris Cedex 05 (France)

Development of our understanding of population genetics, including the analysis of populations on the molecular and biochemical levels, and the rapid growth of molecular biology and biochemistry in recent years, allow a new and more accurate approach to evolution mechanisms, as is demonstrated by Ayala's and Stebbins' articles. The question is then whether the 'Modern Synthesis' theory of evolution, elaborated in the 1940s on a basis of Darwinism, has broken down under the enormous amount of information collected during the last 40 years.

The fundamental claim of Modern Synthesis is that, during geological history, higher taxa diverged gradually, and the mechanisms involved were the same as those we can observe every day in populations. Modern Synthesis, using advances in formal and population genetics, has based evolution on the mutation-selection-drift triptych. Selection, working on the variability of populations, progressively adapts genetic pools to environments. Since available environments on earth are discontinuous, a number of genetic pools are isolated from each other by speciation (microevolution), then diverge more and more in higher taxa (macroevolution), the mechanisms being the same in micro and macroevolution[1–4].

For Darwin, the main difficulty was the lack of knowledge about the nature of variability; for us, the main difficulty lies in the enormous amount of biochemical polymorphism demonstrated first by electrophoresis and, today, by the modern techniques of DNA analysis. While Lewontin[5] in 1966 wrote: 'We then have a dilemma', Ayala can write in this review (p. 820): 'If the non-coding regions of genes are included, it seems likely that most, if not all genes are heterozygous in every outbred individual', which makes one feel dizzy, and raises the question of why such a high level of polymorphism has been maintained in spite of selection.