New estimates of asymmetric decomposition of racemic mixtures by natural β-radiation sources

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The Vester–Ulbricht hypothesis suggests that the chirality of biological molecules originates from the β-radiolysis of prebiotic racemic mixtures[1]. Despite the inconclusiveness of past investigations[2–4], recent calculations[5,6] have shown that β particles, because of their helicity, radiolyse L- and D-enantiomers at slightly different rates, the asymmetry, A_R, being predicted to be 10^{-11} (new experimental tests[7,8] give $|A_R| < 2\times10^{-9}$). Before this, the size of the radiolysis-induced chiral polarization, η_R ($\eta \equiv (n_L - n_D)/(n_L + n_D)$ where n_L and n_D are the numbers of L and D molecules present), was estimated[9–12] for different values of A_R; according to Keszthelyi *et al.*[9–11], if $|A_R| \sim 10^{-11}$, $|\eta_R|$ can never exceed the chiral polarization, $|\eta_F|$, produced by statistical fluctuations, thus invalidating the V–U hypothesis. Here we re-examine the major assumptions on which these calculations were based and find that several overly restrictive conditions were imposed, which, when relaxed, allow the condition $|\eta_R| > |\eta_F|$, in accordance with the V–U hypothesis.

The previous calculations[9–12] of η_R were made for a particular system of chiral molecules undergoing simultaneous formation, β radiolysis, thermal- and radio-racemization and decomposition and, in some[9–11], well-defined physical restrictions were placed on the volume and surface area of the system. We follow here these earlier treatments, except that the restrictions are removed and three new features introduced: (1) the recent theoretical estimate $A_R \sim 10^{-11}$ is used; (2) a number of plausible new intense sources of β radiation are considered (Table 1); (3) the temperature dependence of the racemization and decomposition rate constants is included.

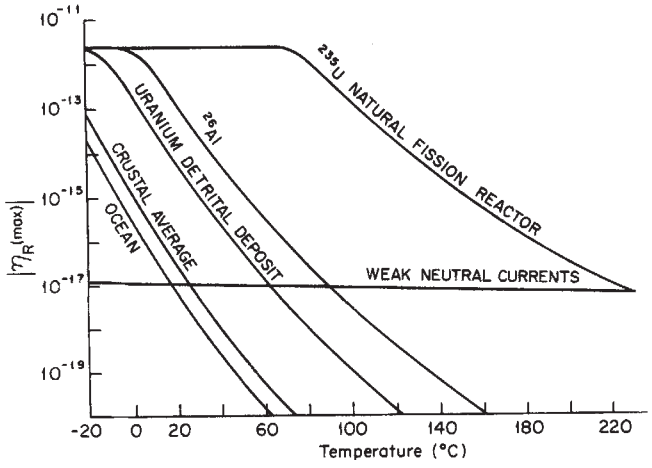

Fig. 1 Magnitude of maximum chiral polarization $|\eta_R(max)|$ versus temperature for radiolysis of alanine in an aqueous environment by natural sources of β radiation. Calculations used the rate equations of Mann and Primakoff[12]. Radiolysis rate constants, for which the asymmetry is 10^{-11} (refs 5, 6), are given in Table 1. The Arrhenius parameters from which the thermal racemization rate constants are calculated, the thermal decomposition rate constants and the radio-racemization rate constant were obtained from refs 27, 28 and 12, respectively. Clearly the largest values of $|\eta_R(max)|$ are favoured by strong β emitters and by low temperatures (the temperature dependence of $|\eta_R(max)|$ is a direct consequence of that of the racemization rate constant). Also shown for comparison is the chiral polarization produced by weak neutral currents, which create an energy difference between an enantiomer and its mirror image[29].

Table 1 Source strengths and radiolysis rate constants for β-ray sources

β-ray source	Source strength $S(s^{-1}\,cm^{-3})$	Radiolysis rate constant $\varepsilon S(s^{-1})$	Radiolysis time constant $(\varepsilon S)^{-1}$ (yr)
^{40}K, ^{14}C in ocean	0.2	5×10^{-19}	6×10^{10}
Total crustal average	3.9	2×10^{-18}	2×10^{10}
Uranium-detrital deposit	6×10^{2}	4×10^{-16}	8×10^{7}
^{26}Al	1×10^{4}	1×10^{-14}	3×10^{6}
^{235}U natural reactor	1.4×10^{9}	5×10^{-10}	60

The estimated source strengths and calculated values of the radiolysis rate constant and radiolysis time constant for several sources that may have occurred where the amino acids and sugars used in the formation of life were present. The values for the ocean and Earth's crust are taken from abundances of ^{40}K, ^{238}U and ^{235}U in ref. 18. In addition to these widespread sources, concentrated sources of radioactivity may have occurred in locations considered favourable to the formation of life—the most probable sources on the prebiotic earth being detrital uranium deposits (E. W. Heinrich, personal communication) occurring along beaches and in streams[19]. Characteristic source strengths for such deposits are taken from Hiemstra[20]. In local areas, uranium concentrations as high as 10% can occur[20], possibly leading to the formation of natural nuclear reactors, such as at Oklo 2×10^9 yr ago[21]. The β intensity for a 10 kW reactor is taken from Lamarsh[22]. Finally, it has been speculated[23] that the amino acids present on the early Earth were formed in the presolar nebula and carried down in carbonaceous chondrites in the last stages of the Earth's accretion. These amino acids would have been irradiated during the decay of nova[24] or supernova[25] produced short-lived radioisotopes incorporated directly into the carbonaceous chondrites, for example, ^{26}Al, which has been detected in the Murchison and other meteorites[26]. The ^{26}Al source strength is obtained from data in ref. 26.

The result of our calculation is that, for a system of N chiral molecules ($N \equiv n_L + n_D$), $|\eta_R|$ increases with time from zero to a maximum value given approximately by

$$\eta_R(\max) \cong \begin{cases} \frac{1}{4}A_R & \text{if } \varepsilon S \gtrsim \beta \\ \varepsilon S A_R/2\beta & \text{if } \varepsilon S \ll \beta \end{cases}$$

where εS is the radiolysis rate constant for a β-ray source of strength S (Table 1) and β is the racemization rate constant or half the thermal-decomposition rate constant, whichever is greater. This result is obtained by solving the rate equations[12] for η_L and η_D to first order in A_R. This maximum value is attained in a characteristic time (defined as the inverse of the respective rate constant) approximately equal to the radiolysis, racemization, or thermal decomposition time, whichever is least. These results are obtained irrespective of whether formation of the enantiomers is taken to occur before, or simultaneously with, radiolysis. For each of the β-ray sources, $|\eta_R(max)|$ is plotted for the amino acid alanine as a function of temperature (Fig. 1) and the corresponding time required to reach this maximum is given for selected temperatures in Table 2. Because of uncertainties in the input parameters, these values are estimates of an order of magnitude rather than precise predictions.

We find that $|\eta_R|$ never exceeds 3×10^{-12}; this is much smaller than previous estimates[12]. Hence, asymmetric β-radiolysis can never by itself produce $|\eta| = 1$. Nevertheless, the small value of η_R can be amplified by subsequent chemical or biological processes eventually to give $\eta = \pm1$. A necessary condition for the validity of the V–U hypothesis is that this initial η_R^* be greater in magnitude than $|\eta_F|$ and, because, from statistical arguments, $|\eta_F|$ is almost certainly $< 3N^{-1/2}$ (this equals three standard deviations, assuming the distribution of fluctuations to be gaussian), this condition will be satisfied for $N > 9/\eta_R^2$. For instance, if $\eta_R = 3\times10^{-12}$, then $N \sim 10^{24}$ (150 g of alanine) which, for reasons to be discussed below, may not be an excessively large amount.

Therefore, for plausible prebiotic conditions, the relation $|\eta_R| \gtrsim |\eta_F|$ can obtain, whereas the most recent analysis of Keszthelyi[13] concludes that $|\eta_R| < |\eta_F|$. The discrepancy arises because Keszthelyi's analysis is based on the choice of a system having a static 1-litre volume with a 1-cm^2 surface area and 10-m depth, which is radiolysed by β particles from natural ^{40}K at a temperature $T \cong 20\,°C$[9–11,13], and assumes that the amino acids produced in the atmosphere enter this volume only through the surface, at a rate of $2\times10^{10}\,s^{-1}\,cm^{-2}$. According to the results

Table 2 Maximum chiral polarization times and magnitudes

β-Ray source	Temperature (°C)	Maximum chiral polarization $\lvert\eta_R\,(\max)\rvert$	Time required (Yr)
^{40}K, ^{14}C in ocean	−20	2×10^{-14}	2×10^{8}
	0	3×10^{-16}	4×10^{6}
	20	7×10^{-18}	9×10^{4}
	100	1×10^{-22}	2
Total crustal average	−20	8×10^{-14}	2×10^{8}
	0	1×10^{-15}	4×10^{6}
	20	3×10^{-17}	9×10^{4}
	100	5×10^{-22}	2
Uranium-detrital deposit	−20	3×10^{-12}	8×10^{7}
	0	2×10^{-13}	4×10^{6}
	20	5×10^{-15}	9×10^{4}
	100	1×10^{-19}	2
^{26}Al	−20	3×10^{-12}	3×10^{6}
	0	3×10^{-12}	3×10^{6}
	20	1×10^{-13}	9×10^{4}
	100	3×10^{-18}	2
^{235}U reactor (10 kW)	−20	3×10^{-12}	60
	0	3×10^{-12}	60
	20	3×10^{-12}	60
	100	1×10^{-13}	2
	350	2×10^{-20}	2×10^{-7}

in Fig. 1, the largest value of $|\eta_R|$ that can be attained under these conditions is $\sim10^{-17}$, requiring $N \geq 10^{35}$ for the condition $|\eta_F| \leq |\eta_R|$ to obtain. However, the number of amino acids accumulating in the system of Keszthelyi is, at most, of order 10^{27} (attained in 10^9 yr), which gives $|\eta_F| \cong 10^{-13}$ and $|\eta_F| > |\eta_R|$.

Figure 1 shows, however, that $|\eta_R| \geq |\eta_F| \cong 10^{-13}$ can obtain by increasing $|\eta_R|$ for the above system, either if $T < -20$ °C, or if the stronger sources that we have introduced are present. In addition $|\eta_F|$ can be smaller than 10^{-13} if larger systems are allowed, for example, by increasing the 1-cm^2 surface area of the systems in refs 9–11, 13. We note that subsequent amplification mechanisms may well impose limits on the size of a system, where 'system' is defined as the total number of chiral molecules, N, which is the starting point for chemical or biological processes amplifying $|\eta|$ in that initial population. However, the nature of such processes has not been determined explicitly, although models amplifying $|\eta|$ in systems of large physical dimensions have been demonstrated mathematically[14–16]. Thus, we conclude that there is no justification for assuming $N < 10^{27}$ as in refs 9–11.

We have demonstrated that, under certain not improbable conditions, the result $|\eta_R| \geq |\eta_F|$ can obtain despite the small magnitude of η_R. Recently, a general argument that some specific causal mechanism probably dominated statistical fluctuations has been advanced[17], which our results show could have been asymmetric radiolysis. Considering the present lack of knowledge of the details of any realistic prebiotic amplification process and evidence presented here for the prebiotic existence of the strong ^{235}U and ^{26}Al sources (Table 1), the V–U hypothesis cannot be excluded without further detailed research.

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Cyclic AMP-dependent protein kinase closes the serotonin-sensitive K^+ channels of *Aplysia* sensory neurones in cell-free membrane patches

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Selected actions of neurotransmitters and hormones on ion channels in nerve and muscle cells are now thought to be mediated by cyclic AMP-dependent protein phosphorylation[1–11]. Although the cyclic AMP-dependent protein kinase (cAMP-PK) affects the cellular properties of several neurones, its mode of action at the single-channel level has not been characterized. In addition, little is known about the identity or subcellular localization of the phosphoproteins that control channel activity and, in particular, whether the critical substrate proteins are cytoplasmic or membrane-associated. In *Aplysia* sensory neurones, serotonin produces a slow modulatory synaptic potential mediated by cAMP-PK[12] that contributes to presynaptic facilitation and behavioural sensitization[12]. Previously, we have found that serotonin acts on cell-attached membrane patches to produce prolonged all-or-none closures of a specific class of K^+ channels (S channels) whose gating is weakly dependent on voltage and independent of intracellular calcium[13–15]. We demonstrate here that in cell-free membrane patches from *Aplysia* sensory neurones, the purified catalytic subunit of cAMP-PK[16] produces all-or-none closures of the S channel, simulating most (but not all) aspects of the action of serotonin on cell-attached patches. This result suggests that protein kinase acts on the internal surface of the membrane to phosphorylate either the channel itself or a membrane-associated protein that regulates channel activity.

Single-channel currents were recorded from cell-free inside-out patches of membrane (cytoplasmic surface of the membrane facing the bath) from LE sensory neurones of *Aplysia* abdominal ganglia using standard techniques[13,17]. The membrane potential across the patch was held at depolarized levels to inactivate both the delayed rectifier K^+ channel and the early K^+ channel[18]. In these conditions, the two types of channel currents observed in the cell-free patches corresponded to the non-inactivating S-channel current (I_s) and a Ca^{2+}-activated K^+-channel current (I_c)[18,19]. The S channel in cell-free patches displays properties similar to those seen in voltage-clamp[14] and cell-attached patch-clamp experiments[13,15]. To study the action of cAMP-PK on S-channel currents in isolation, we kept the concentration of Ca^{2+} in the bath at 0.2 μM, a concentration which does not activate the Ca^{2+}-activated K^+ channel in *Aplysia* at the potential at which the patch was held.

In experiments with cAMP-PK, S-channel currents were first identified in the inside-out patch on the basis of their Ca^{2+}-insensitivity and unit current amplitude. The patch pipette was