

# GRADED AUTOCATALYSIS REPLICATION DOMAIN (GARD): KINETIC ANALYSIS OF SELF-REPLICATION IN MUTUALLY CATALYTIC SETS

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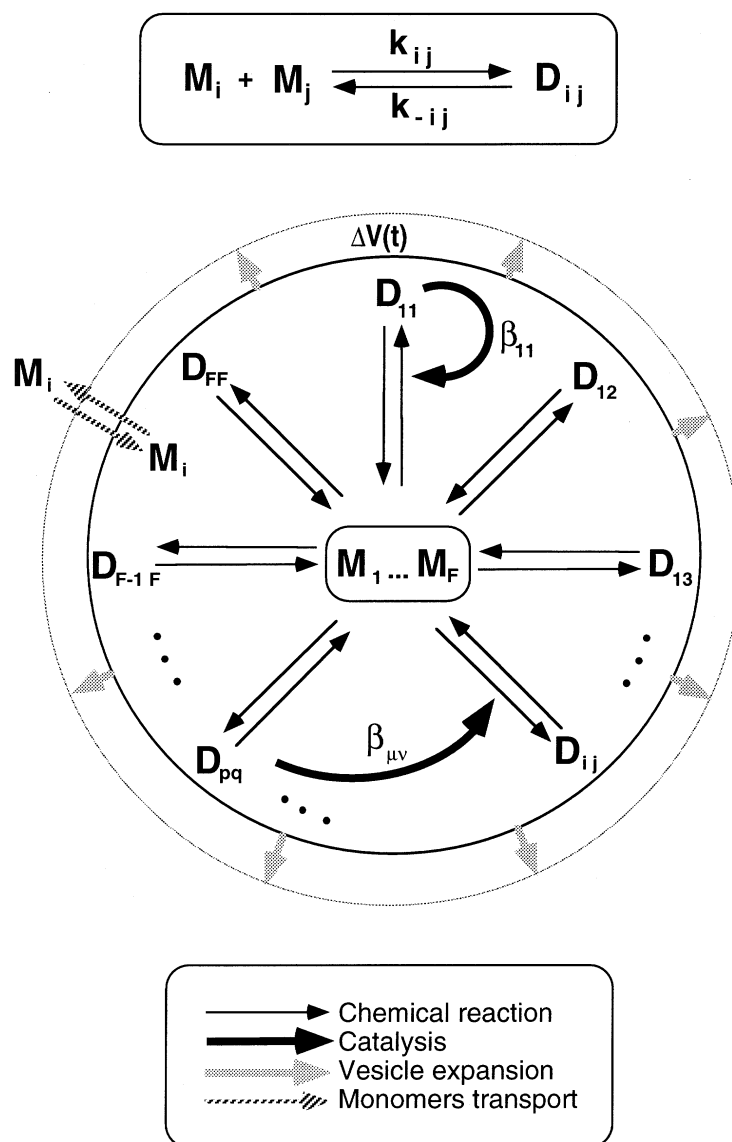
**Abstract.** A Graded Autocatalysis Replication Domain (GARD) model is proposed, which provides a rigorous kinetic analysis of simple chemical sets that manifest mutual catalysis. It is shown that catalytic closure can sustain self-replication up to a critical dilution rate,  $\lambda_c$ , related to the graded extent of mutual catalysis. We explore the behavior of vesicles containing GARD species whose mutual catalysis is governed by a previously published statistical distribution. In the population thus generated, some GARD vesicles display a significantly higher replication efficiency than most others. GARD thus represents a simple model for primordial chemical selection of mutually catalytic sets.

## 1. Introduction

Primordial evolution requires self-replication of chemical species. Autocatalysis and template-mediated replication of individual molecules have been pursued as the basis for such a process (Eigen, 1971; Swetina and Schuster, 1982; Küppers, 1983; Lifson, 1987; Orgel, 1992; Li and Nicolaou, 1994; Siever and Von Kiedrowski, 1994). In parallel, it has been suggested (Dyson, 1985; Farmer, Kauffman *et al.*, 1986; Kauffman, 1993; Stadler, Fontana *et al.*, 1993; Fontana and Buss, 1994) that sets of biopolymers with mutual catalysis may undergo replication (due to 'catalytic closure' (Farmer, Kauffman *et al.*, 1986; Kauffman, 1993)), even if none of the individual components is autocatalytic. The latter scenario has been proposed to represent a primitive self-propagating metabolism without a genome (cf. (Wächtershäuser, 1990; Kauffman, 1993)). We describe here a kinetic model which provides a quantitative measure of self-replication of species in mutually catalytic sets. The results address a potential scenario for rudimentary replication behavior in a primordial, very heterogeneous mixture of organic molecules (Miller, 1953), members of which may be expected to display weak catalysis. Such a scenario could predate a much later stage, at which self-replication began to be performed by more complex information-carrying or catalytic biopolymers (Eigen, 1971; Orgel, 1992).

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*Figure 1.* A schematic depiction of the GARD model. The term GARD (Graded Autocatalysis Replication Domain) denotes a collection of  $N$  types of organic molecules, that are chemically interconvertible via common precursors, and are contained in a spatial domain with a defined volume. GARD is governed by graded mutual catalysis and its kinetic properties resemble those of a replicating molecular autocatalyst. A GARD vesicle is shown, containing the high energy monomers  $M_i$ , and their dimers  $D_{ij}$ . They interconvert by reversible chemical reactions with their respective kinetic constants (top box). The catalysis arrows connecting  $D_{pq}$  with the reaction  $M_i + M_j \rightleftharpoons D_{ij}$  represent the rate enhancement effect of  $D_{pq}$  on the formation and degradation of  $D_{ij}$  with a multiplicative factor  $\beta_{\mu\nu} \equiv \beta_{\mu(i,j)\nu(p,q)}$ . The monomer transport arrows represent the inward and outward free passive diffusion of the ‘food set’ materials  $M_i$  which are buffered across the vesicular wall. The vesicle expansion arrows indicate the volume increment  $\Delta V(t)$  in the time interval  $\Delta t$ .

## 2. Results

We consider here a simplified representation of a catalytic set (cf. (Kauffman, 1993)) as shown in Figure 1. In this embodiment, which is more realistic than those we analyzed before (Lancet, Kedem *et al.*, 1994; Lancet, Glusman *et al.*, 1995; Lancet, Glusmann *et al.*, 1996; Segré, Pilpel *et al.*, 1996)  $N_G F^2$  dimeric species,  $D_{ij}$ , are generated from a set of  $F$  high energy monomers  $M_1, M_2, \dots, M_F$ , supplied from outside the system ('food set' (Kauffman, 1993)). This takes place in a set of reversible bimolecular reactions with rate constants  $k_{ij}, k_{-ij}$ . The specific model considered here is useful for analyzing the most fundamental features of mutual catalysis and catalytic closure. More complex embodiments, with more comprehensive reaction topologies have been proposed (Farmer, Kauffman *et al.*, 1986; Kauffman, 1993; Stadler, Fontana *et al.*, 1993; Fontana and Buss, 1994) and could be used to extract additional information in the future.

In previous analyses (Farmer, Kauffman *et al.*, 1986; Kauffman, 1993), each species within the catalytic set (symbolizing any organic compound, including amino acids, nucleotides and their oligomers) was deemed as a potential catalyst for one of the system's chemical reactions. The present model considers a generalization, in which mutual catalysis is defined by an  $N_G \times N_G$  matrix  $\beta$ , whose element  $\beta_{\mu\nu} \equiv \beta_{\mu(i,j)\nu(p,q)}$  represents the catalytic enhancement factor of the species  $D_{pq}$  on the reaction  $M_i + M_j \rightleftharpoons D_{ij}$  ( $1 \leq \mu, \nu \leq N_G$ ). In this notation, diagonal  $\beta$  matrix elements signify autocatalysis, and off-diagonal elements represent mutual catalytic events.

Applying the basic laws of chemical kinetics, it can be seen that the time dependent concentration of the species  $D_{ij}$  obeys the differential equation:

$$\begin{aligned} \frac{dD_{ij}}{dt} = & k_{ij}M_iM_j - k_{-ij}D_{ij} + \\ & + k_{ij}M_iM_j \sum_{p,q=1}^F \beta_{\mu(i,j)\nu(p,q)}D_{pq} - k_{-ij}D_{ij} \sum_{p,q=1}^F \beta_{\mu(i,j)\nu(p,q)}D_{pq}. \end{aligned} \quad (1)$$

The first two terms in Equation 1 represent the uncatalyzed (spontaneous) formation and decomposition of  $D_{ij}$ , that are expected to be very slow. The last two terms represent the mutually catalyzed forward and backward reactions. In reality, most  $\beta_{\mu\nu}$  may be extremely small and only rarely will appreciable  $\beta_{\mu\nu}$  appear, as expected for random interactions between organic molecules, including enzyme mimetics (Bar-Nun, Kochavi *et al.*, 1994; Kirby, 1994).

A central question to be addressed in this paper is what might be the kinetic properties of such a set of chemicals, which would allow it to undergo self-replication. For this, we consider a realistic embodiment, in which a subset of  $N$  out of the  $N_G$  possible  $D_{ij}$  components is spatially constrained, e.g. by enclosure within a membrane vesicle (cf. (Bachmann, Luisi *et al.*, 1992; Pohorille and Wilson, 1995)).

The wall of such vesicle is assumed to be impermeable to all the  $D_{ij}$  species, but to pass freely the food set monomers  $M_i$ . The monomer concentrations  $M_i$  are thus kept constant within the vesicle through fast equilibration with a large external pool. Self-replication may then be defined as a process that, through mutual catalysis, provides for making additional copies of all the enclosed molecular species  $D_{ij}$  from the monomeric precursors  $M_i, M_j$ . We name the vesicle-enclosed mutually-catalytic set GARD: Graded Autocatalysis Replication Domain.

It is possible to consider a process in which the volume available for the GARD's components increases 2 fold, while all concentrations  $D_{ij}$  are preserved by further synthesis. In this process the copy number of all molecules is doubled. If the GARD subsequently splits into two separate volumes, a simple replication process is seen to have occurred. As shown below, this may happen under steady state conditions, far from equilibrium, whereby a conjectural composition preserved by mutual catalysis is being propagated. Thus, it is suggested that self-replication in a set of interconvertible chemicals, such as represented in a GARD, is equivalent to the homeostatic preservation of the steady state attained under conditions of persistent dilution. This is analogous to previous definitions of replicative steady states (Eigen, 1971) except that here the individual components are not assumed to be self-replicating information carriers, and information is manifested in the composition of the set as a whole.

If the available volume in a GARD increases exponentially, as would be the case for vesicles that expand and undergo splitting (Rashevsky, 1960; Bachmann, Luisi *et al.*, 1992; Luisi, Walde *et al.* 1994), then  $V(t) = V(0) \exp(\lambda t)$ , and Equation (1) becomes (see appendix A):

$$\frac{dD_{ij}}{dt} = [k_{ij}M_iM_j - k_{-ij}D_{ij}] \left[ 1 + \sum_{p,q=1}^F \beta_{\mu(i,j)\nu(p,q)} D_{pq} \right] - \lambda D_{ij}. \quad (2)$$

The balance between the reactions and the dilution expressed in Equation (2) is rather general, and may also refer to mechanisms other than vesicle enclosure, such as desorption in a set of surface-adsorbed chemicals. Equation (2) predicts a steady state, which depends on the balance between the decline of all  $D_{ij}$  due to expansion-related dilution, and the (potentially catalyzed) generation of each  $D_{ij}$ . Numerical solution of Equation (2) indicates a single steady state if all kinetic constants are positive, while more complex time dependencies may appear otherwise (Goldbeter and Nicolis, 1972).

Figures 2a–d show numerical solutions of Equation (2) with different values of  $\lambda$ . This is done for a very simple GARD with  $F=3$  and  $N_G = N = 9$ . The aim of this analysis is to examine the specific effect of mutual catalysis on the time-dependent differences in the  $D_{ij}$  concentrations. This is best done by analyzing species  $D_{ij}$  that are equally favorable thermodynamically ( $K_{ij}=k_{ij}/k_{-ij}$  equal for all  $ij$ ). For the same reason, we also assume that all  $D_{ij}$  have identical basal kinetic constants

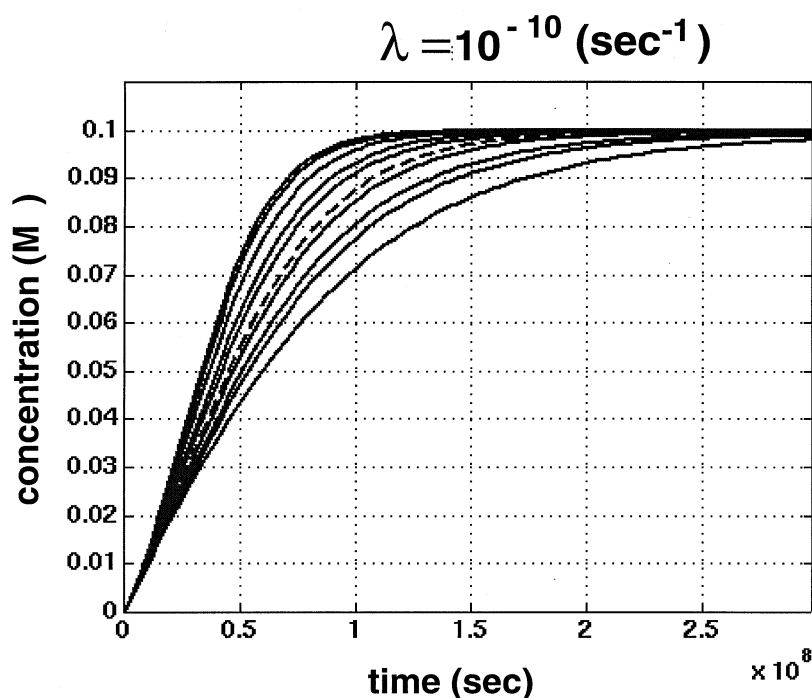


Figure 2a–d. The kinetic behavior of GARD species. All the results shown here and in the next figure were obtained using the GRIND software (De Boer, 1983) for the numerical solution of differential equations, and the NAG Fortran subroutine package for computing steady state values, all done on a Silicon Graphics Indy 2 computer. In a large number of numerical experiments only solutions of Equation (2) that lead to a single steady state with all  $D_{ij} > 0$  were obtained, in accordance with previous analyses (Stadler, Fontana *et al.*, 1993; Schlosser and Feinberg, 1994). The parameter values used throughout this work are:  $k_{ij} = 0.1 \text{ sec}^{-1} \text{ M}^{-1}$ ,  $k_{-ij} = 1 \times 10^{-8} \text{ sec}^{-1}$  ( $K_{ij} = 1 \times 10^7 \text{ M}^{-1}$  representing a considerable free energy change appropriate for the dimerization of high energy monomers),  $M_i = 1 \times 10^{-4} \text{ M}$  for all  $i, j$  values. The time dependent concentrations of the components of a GARD system with  $N_G = N = 9$ , is computed by numerical solution of Equation (2) with varying rates of expansion as noted above each sub-figure. The initial concentration of all  $D_{ij}$  is equal to zero. The elements of the  $\beta$  matrix were selected as shown in sub-figure 2E. The time-dependences of the concentrations of all  $D_{ij}$  are shown by continuous lines. Each dimer has a different time course and a different steady state plateau, depending on the degree of mutual catalysis exerted on it by other GARD species. For  $\lambda = 0$  the steady state concentrations of all species are identical, a situation approximated by the small  $\lambda$  value of sub-figure 2A. The dashed lines depict the kinetic behavior of a single autocatalyst with  $\beta_{11} = 22 \text{ M}^{-1}$ .

( $k_{ij}, k_{-ij}$  are equal for all  $ij$ ). Thus, the only difference among  $D_{ij}$  in this analysis is assumed to be in their respective  $\beta_{\mu\nu}$ .

The selection of the elements  $\beta_{\mu\nu}$  is a crucial part of the model. In an arbitrary collection of dimers  $D_{ij}$  there is no specific information on any of the  $\beta_{\mu\nu}$  values. However, it may be possible to have some knowledge on the statistical distribution out of which  $\beta_{\mu\nu}$  values might be drawn. We have previously proposed a probabilistic Receptor Affinity Distribution (RAD) model for the statistics of affinities

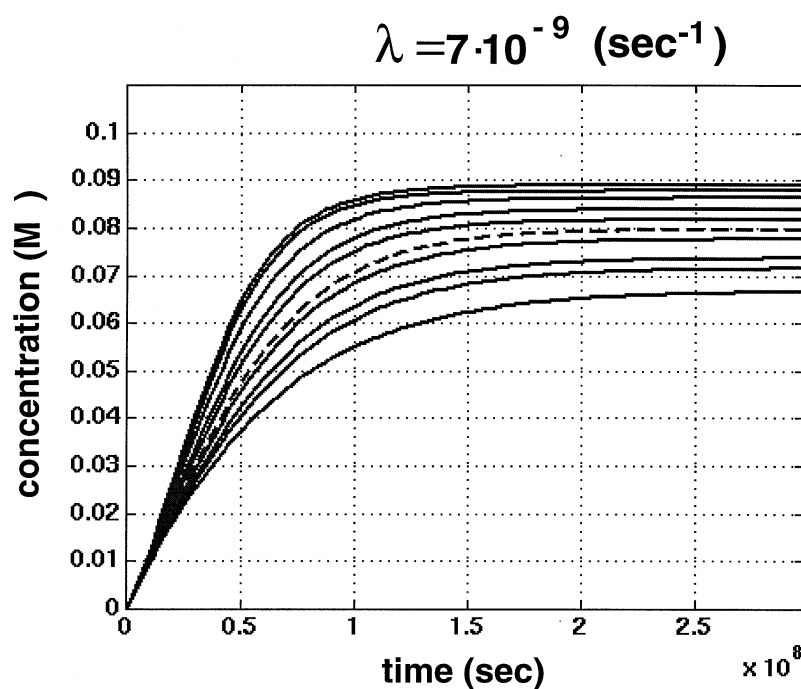


Figure 2b.

in ligand/receptor systems (Lancet, Sadvovsky *et al.*, 1993) and suggested that a similar model may apply also to catalytic enhancement values in enzyme mimetic systems (Lancet, Kedem *et al.*, 1994; Segré, Pilpel *et al.*, 1996). The 9X9  $\beta$  matrix used in Figures 2a–d is derived based on such a probability distribution (Figure 2e and legend).

As seen in Figure 2a, when  $\lambda$  is vanishingly small, the only effect of mutual catalysis is in generating different transients for different  $D_{ij}$ . At longer times all  $D_{ij}$  tend towards the same asymptotic concentration. As the expansion parameter  $\lambda$  increases (Figures 2b,c), each of the species assumes a different steady state concentration, with species that enjoy better mutual catalysis (accumulated over all potential catalysts), having a higher concentration. Finally, as  $\lambda$  is increased further (Figure 2d), none of the species can withstand effectively the dilution due to expansion, and all  $D_{ij}$  concentrations become rather small. These results indicate that, under conditions of persistent dilution, catalysis may change the steady state concentration of thermodynamically identical species. In other words, dilution leads to a unique steady state composition, which may be very different from the equilibrium state, at which all  $D_{ij}$  have equal concentrations. It should be pointed out that the kinetic behavior of a single autocatalyst is mathematically very similar to that shown for the mutually catalytic species (broken line in Figures 2a–d).

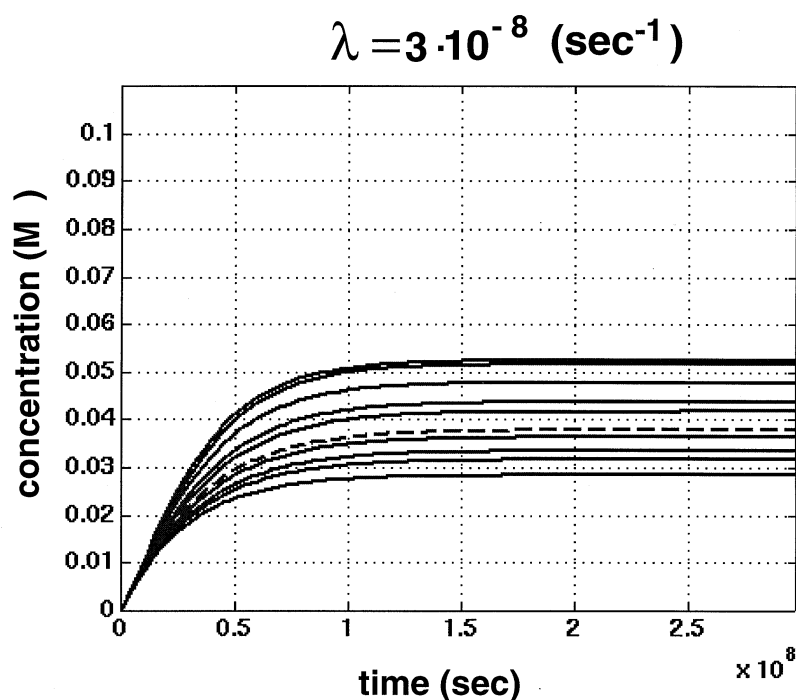


Figure 2c.

The introduction of  $\lambda$  as an externally imposed expansion rate parameter allows one to quantify the self-replication capacity of each chemical species within the GARD. For continuously increasing values of  $\lambda$ , each of the  $D_{ij}$  species will show a different behavior, which depends on the extent of mutual catalysis exerted upon it (Figure 2f). It is possible to define  $\lambda_c$  as the critical value of  $\lambda$  at which the steady state concentration for none of the species  $D_{ij}$  drops below  $1/e$  of its value at  $\lambda=0$  (no dilution). We suggest that  $\lambda_c$  could serve as a quantitative, graded measure for the self-replication capacity of an entire catalytic set (see also (Lancet, Glusman *et al.*, 1995; Lancet, Glusmann *et al.*, 1996; Segré, Pilpel *et al.*, 1996)).

We next consider a more elaborate chemistry with  $N_G = 100$ , using a  $\beta$  matrix of  $100 \times 100$  (Figure 3a). We refer to a random collection of many small GARD vesicles, each initialized with  $n \ll N_G$  dimer molecules. Probability considerations dictate that most such vesicles will have one copy of each of  $n$  kinds, randomly selected, i.e.  $N = n$ . It is then possible to obtain a rough estimate of the catalytic ‘quality’ of such GARD vesicles by computing the steady state concentrations of all species. This is done with the simplifying assumption that no species other than the initial  $n$  will form during the process of steady state attainment.

In order to obtain a view of the statistics of all the possible vesicle types, a total of  $N_G! / [(N_G - N)! N!]$ , we analyze a sample of 10 000 different GARD vesi-

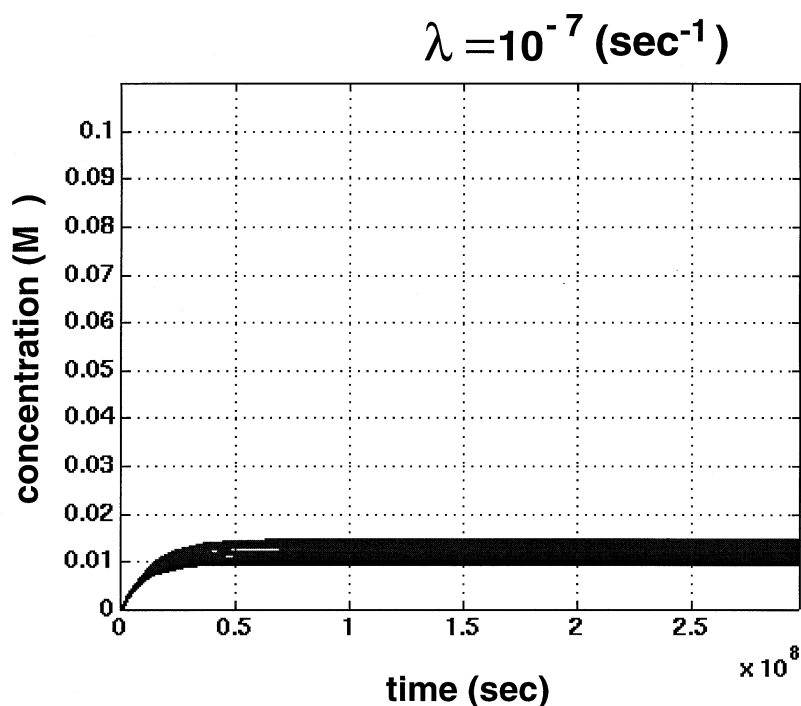


Figure 2d.

cles with  $N=20$ . We choose vesicle expansion characterized by  $\lambda=2 \times 10^{-8} \text{sec}^{-1}$ , assumed to be determined by physicochemical properties of the vesicle wall material (Bachmann, Luisi *et al.*, 1992). For each vesicle we compute the steady state concentrations for all species and take their average. It can be seen that the GARD vesicles show a considerable variation, depicted in the distribution of Figure 3b. The width of such a distribution will depend on the ratio  $q = n/N_G$ : for  $q \gg 1$  there will be many copies of all of the  $N_G$  species in each GARD vesicle, and variation will be negligible. On the other hand, for  $q \ll 1$ , smaller  $n$  values will result, up to a point, in a broader distribution. We are currently investigating the possible existence of an optimal  $q$ , for which the highest variation arises, and where the best GARD vesicles might potentially be generated.

### 3. Discussion

We describe a Graded Autocatalysis Replication Domain (GARD) model for mutual catalysis in a set of simple reversible reactions that occur within an enclosed volume. The model is based on previously developed concepts of mutually catalytic sets and catalytic closure (Farmer, Kauffman *et al.*, 1986; Kauffman, 1993). It is applicable to a time window in chemical evolution in which heterogeneous



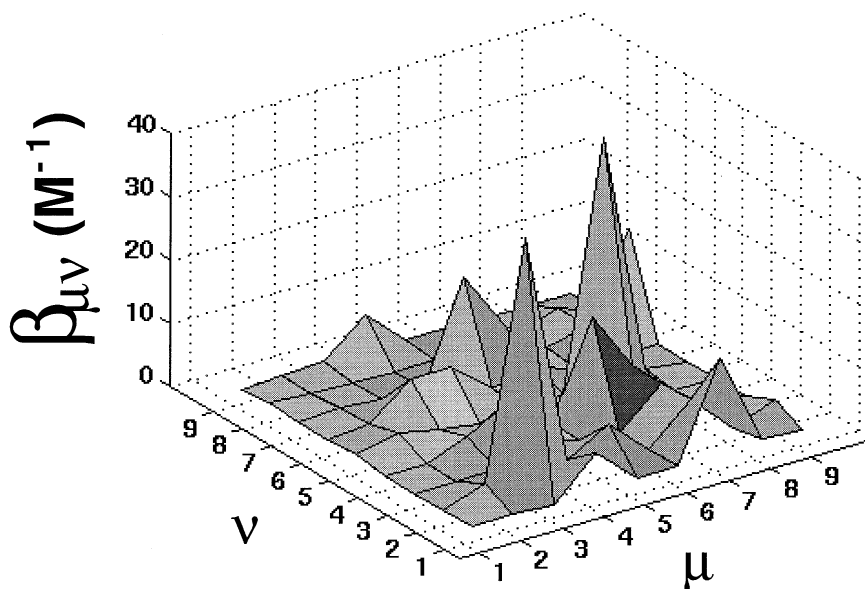


Figure 2e. A graphic representation of a 9X9  $\beta$  matrix derived by random sampling of  $\beta_{\mu\nu}$  values from a binomial probability distribution, as previously proposed (Lancet, Kedem *et al.*, 1994; Segré, Pilpel *et al.*, 1996):  $\Phi(\beta_{ij}) = \text{Binom}[n, p, C_1 \log(\beta_{\mu\nu}) + C_2]$  with the binomial parameters:  $n = 15$ ,  $p = 0.25$ ,  $C_1 = 2.5$ ,  $C_2 = 4$ .

mixtures of organic molecules have already formed from simple precursors, while complex information carriers or biopolymeric catalysts have not yet appeared. A vital point of the model, similar to previous descriptions for mutual catalysis, is its portraying a system in which the catalysts and the catalyzed entities belong to the same set of molecules. The GARD model further allows a rigorous kinetic analysis of the behavior of the mutually catalytic components, providing a quantitative assessment of their self-replication capacity.

We demonstrate that components of a mutually catalytic set, none of which is necessarily autocatalytic, might resemble in their collective behavior the kinetics of a single autocatalyst. The GARD model provides a natural and simple quantitative definition of self-replication capacity of members of catalytic sets. This is based on a graded critical measure ( $\lambda_c$ ) of the ability of the set's components to maintain their concentrations under dilution. GARDs typically have multiple, though small catalysis events for each of the chemicals, and are thus endowed with a remarkable capacity to withstand mutation-like changes, in which one or a few catalytic events are deleted (Pilpel, Lancet and Segré, in preparation). This is in contrast to other systems, in which a mutation or the deletion of one of the catalytic components could result in a catastrophic deterioration of the effectiveness of an entire catalytic cycle (Swetina and Schuster, 1982).

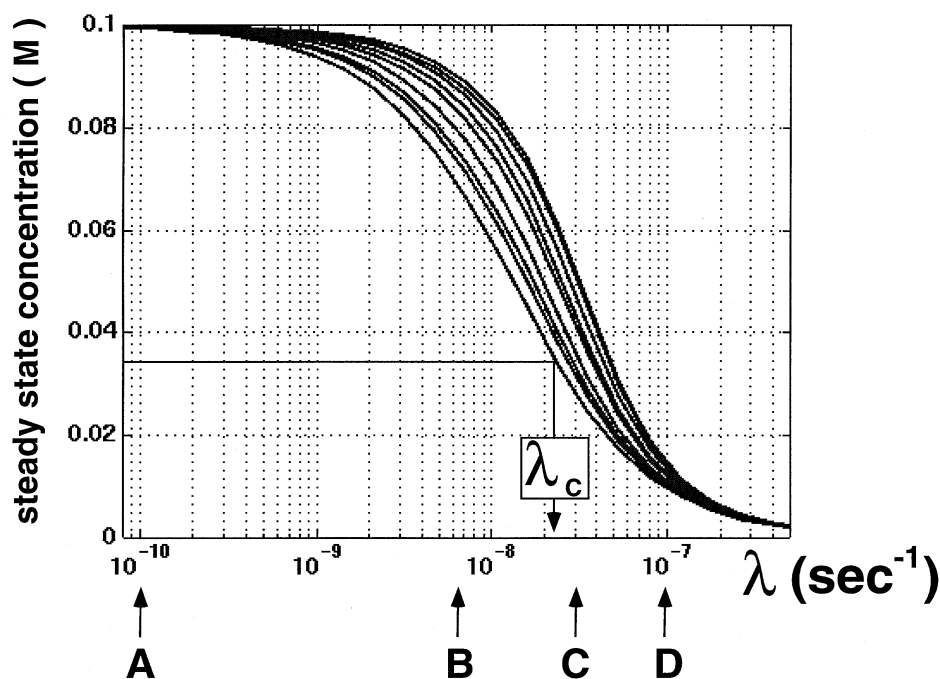


Figure 2f. The dependence of the steady state concentrations of each of the chemical species  $D_{ij}$  on the expansion parameter  $\lambda$ , derived by iteratively solving Equation 2 for steady state.  $\lambda_c$  is that  $\lambda$  at which the steady state concentration for the least catalyzed dimer declines to  $1/e$  of its value at  $\lambda = 0$ . The values of  $\lambda$  used in sub-figures 2A–D are indicated by arrows at the bottom. The concentrations of poorly catalyzed species drop appreciably at relatively small rates of expansion at which other, well catalyzed species can still cope with the dilution effect.

There is a formal similarity between Equation 2 of the GARD model and the kinetic equations of the quasi-species model (Eigen, 1971; Eigen and Schuster, 1979). Both describe multiple species generated from high energy precursors and undergoing a dilution flux. However, the GARD model (Equation 2) includes, in addition, the forward and backward uncatalyzed reactions and a backward catalyzed reaction. This helps achieve a complete chemical kinetic description of the species involved. This is important for the analysis of early prebiotic systems that may have had only a minor deviation from equilibrium. Furthermore, the two models do not refer to the same chemical system. The quasi-species formalism refers mainly to information carriers that undergo self-replication with errors, while the GARD model applies to much simpler chemicals that catalyse each other's formation from precursors. Still, the partial formal similarity between the two models might hint to general hallmarks applicable to different stages of molecular evolution.

In a previous treatise of mutually catalytic sets (Kauffman, 1993) an analysis has been carried out suggesting that as the number of interacting species increases, the probability increases for each species to be catalyzed by at least one other member

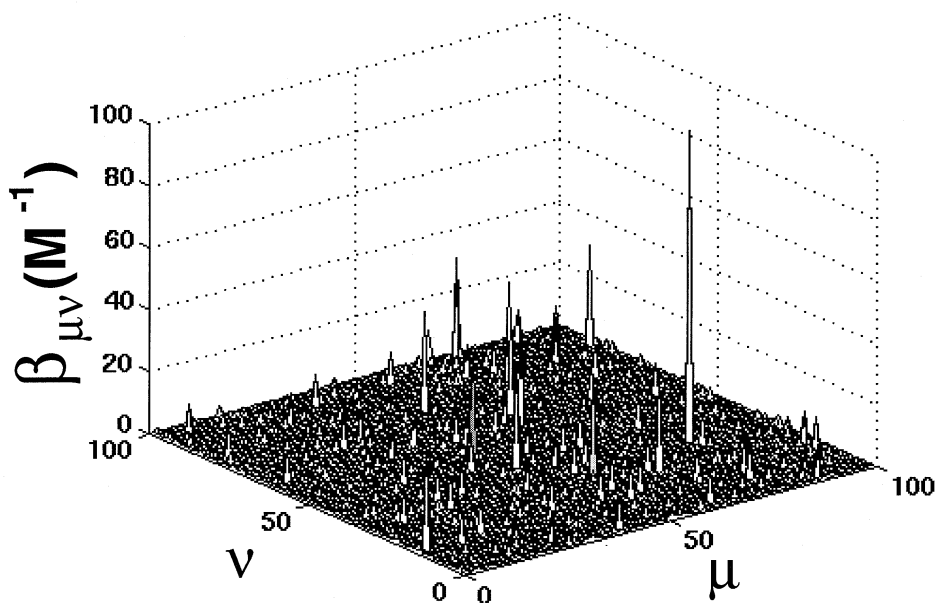


Figure 3a. A graphic representation of an  $N_G \times N_G$   $\beta$  matrix, with  $N_G = 100$ , depicting the entire chemistry governing the GARD vesicles analyzed in sub-figure 3B. The  $\beta_{\mu\nu}$  values were sampled from a distribution function  $\Phi(\beta_{ij})$  with the parameters:  $n = 15$ ,  $p = 0.125$ ,  $C_1 = 2$ ,  $C_2 = 4$ . This was chosen such that most values (>95%) of mutual catalysis are smaller or equal than 1 (i.e. catalyzed rate  $< X2$  the basal rate), as expected for weak catalytic interactions among members of a random chemical set.

of the set. This led to the consideration of very large number of species in a mutually catalytic set. A criticism voiced against this approach has been that as the number of species increases, the network becomes too 'dense'. At the same time, since volume is finite, the concentration of many species may become too small. Such a situation may render the kinetics of the reaction highly ineffective. In the present GARD model we have considered an alternative, whereby each local environment (GARD vesicle) contains only a limited number of species, at manageable concentrations. Optimal mutual catalysis may be reached in a few GARD vesicles that reside at the extreme end of a probability distribution of mutual catalytic effectiveness.

Heterogeneous populations of GARDs, in which some show more efficient mutual catalysis than others, may constitute a simple system for biochemical diversity. GARDs could undergo primordial evolution if the conditions arise that allow those with a higher critical expansion parameter  $\lambda_c$  to expand in correlation with their replication power. This will result in a selective augmentation of the abundance of such GARD vesicles. The analysis of such phenomena is currently underway, using both the approach of solving differential equations, and of stochastic computer simulations.

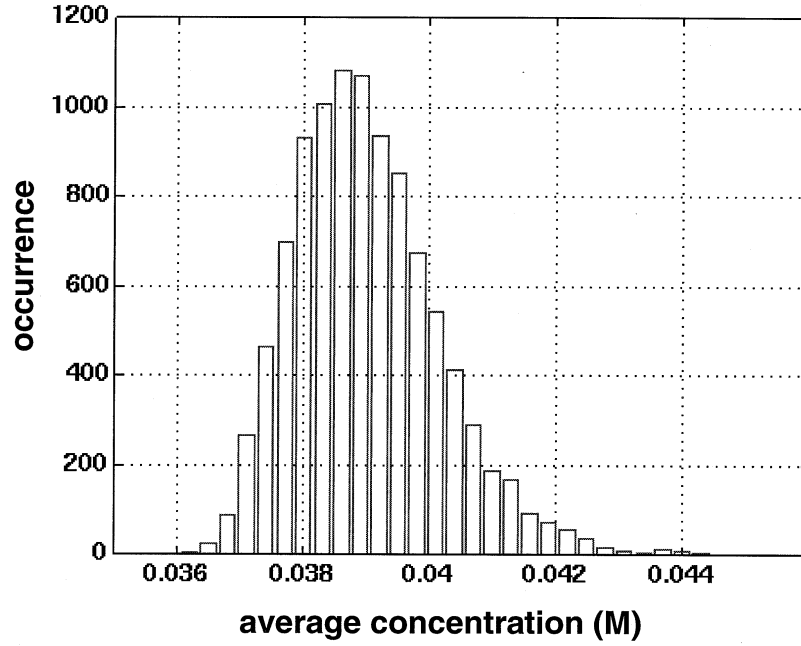


Figure 3b. A frequency histogram of the average molar concentration dimers within GARD vesicles subject to an expansion rate  $\lambda = 2 \times 10^{-8} \text{ sec}^{-1}$ , with all other kinetic constants as in Figure 2. The histogram represents 10000 randomly sampled GARD vesicles, each containing  $N = 20$  species  $D_{ij}$  out of the possible  $N_G = 100$ . For all GARDs the  $N \times N$  mutual catalysis matrix was constructed by sampling from the  $N_G \times N \times N_G$   $\beta$  matrix shown in sub-figure 3A. This distribution is representative of the central region of the distribution expected for the total number of possible GARDs with  $N = 20$ , drawn out of  $N_G = 100$ , i.e.  $100!/(80! \times 20!) \approx 5 \times 10^{20}$ . Thus, the maximal predicted steady state concentration is expected to be much higher than the maximum explored in the partial distribution shown.

## Appendices

A. In an expanding vesicle of volume  $V(t)$ , the time-dependent concentration of the GARD components relates both to the kinetics of the chemical conversions  $M_i + M_j \rightleftharpoons D_{ij}$  (basal and catalyzed), and to the rate of expansion.

In order to obtain Equation 2, we modify Equation 1 introducing a term for the change in concentration due to expansion. Since expansion does not involve a change in the amount of material,  $D_{ij} \times V$  is constant in time and hence (using  $X' \equiv dX/dt$ ):

$$0 = (D_{ij} \times V)' = D_{ij}' \times V + D_{ij} \times V'$$

Therefore

$$D_{ij}' = -(V'/V) \times D_{ij}.$$

If we assume

$$V(t) = V_0 \exp(\lambda \times t),$$

then

$$V' = \lambda \times V_0 \exp(\lambda \times t)$$

and

$$V'/V = \lambda.$$

Thus  $D'_{ij} = -\lambda \times D_{ij}$  is the contribution of the expansion to the derivative of the concentration, to be added to Equation 1 for obtaining Equation 2.

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### References

- Bachmann, P. A., Luisi, P. L. and Lang, J.: 1992, *Nature* **357**, 57.  
 Bar-Nun, A., Kochavi, E. and Bar-Nun, S.: 1994, *J. Mol. Evol.* **39**, 116.  
 De Boer, R. J.: 1983, *GRIND: Great Integrator of Differential Equations*, Utrecht, Utrecht University.  
 Dyson, F.: 1985, *Origins of Life*, Cambridge, Cambridge University Press.  
 Eigen, M.: 1971, *Naturwissenschaften* **58**, 465.  
 Eigen, M. and Schuster, P.: 1979, *The Hypercycle*, Berlin, Springer Verlag.  
 Farmer, J. D., Kauffman, S. A. and Packard, N. H.: 1986, *Physica D* **22**, 50.  
 Fontana, W. and Buss, L. W.: 1994, *Proc. Natl. Acad. Sci. USA* **91**, 757.  
 Goldbeter, A. and Nicolis, G.: 1972, *Biophysic* **8**, 212.  
 Kauffman, S. A.: 1993, *The Origins of Order – Self-Organization and Selection in Evolution*, Oxford, Oxford University Press.  
 Kirby, A. J.: 1994, *Angew. Chem. Int. Ed. Eng* **33**, 551.  
 Küppers, B. O.: 1983, *Molecular Theory of Evolution*, Berlin, Springer.  
 Lancet, D., Glusman, G., Segré, D., Kedem, O. and Pilpel, Y.: 1995, 'A Cellular Automaton Model for Self-Replication of Mutually Catalytic Biopolymers', Proceedings of the 3rd European Conference on Artificial Life., Granada, Spain.  
 Lancet, D., Glusmann, G., Segré, D., Kedem, O. and Pilpel, Y.: 1996, *Origins Life Evol. Biosphere* **26**, 270–271.  
 Lancet, D., Kedem, O. and Pilpel, Y.: 1994, *Ber. Bunsenges. Phys. Chem.* **98**(9), 1166.  
 Lancet, D., Sadovsky, E. and Seidemann, E.: 1993, *Proc. Natl. Acad. USA* **90**, 3715.  
 Li, T. and Nicolaou, K. C.: 1994, *Nature* **369**, 218.  
 Lifson, S.: 1987, *Biophys. Chem.* **26**, 303.  
 Luisi, P. L., Walde, P. and Oberholzer, T.: 1994, *Ber. Bunsenges. Phys. Chem* **98**(9), 1160.  
 Miller, S. L.: 1953, *Science* **117**, 528.  
 Orgel, L. E.: 1992, *Nature* **358**, 203.  
 Pohorille, A. and Wilson, M. A.: 1995, *Origins Life Evol. Biosphere* **25**, 21.

- Rashevsky, N.: 1960, *Mathematical Biophysics: Physico-Mathematical Foundations of Biology*, New York, Dover publications INC.
- Schlosser, P. M. and Feinberg, M.: 1994, *Chem. Eng. Sci.* **49**, 1749.
- Segré, D., Pilpel, Y., Glusman, G. and Lancet, D.: 1996, in C. B. Cosmovici, S. Bowyer and D. Werthimer, *Astronomical and Biochemical Origins and the Search for Life in the Universe*, Bologna, Editrice Compositori, 469–476.
- Siever, D. and Von Kiedrowski, G.: 1994, *Nature* **369**, 221.
- Stadler, P. F., Fontana, W. and Miller, J. H.: 1993, *Physica D* **63**, 378.
- Swetina, J. and Schuster, P.: 1982, *Biophys. Chem.* **16**, 329.
- Wächtershäuser, G.: 1990, *Proc. Natl. Acad. Sci. USA* **87**, 200.