Protocell autonomy:
linking processes through self-made boundaries

Kepa Ruiz-Mirazo

Conference on Bioscience and Society: Interdependence of processes
Ljubljana, 6th-7th Oct. 2011
Universal biochemical features

- Homochirality
- DNA as the genetic material
- Energy currencies
- Cellular boundary
- Common coenzymes and metabolic intermediaries
- Genetic code
LIFE: SYSTEM PROPERTY ! → ORIGINS: SYSTEMIC APPROACH!!

A universal definition of ‘minimal life’

- ‘autonomy’
- ‘open-ended evolution’

CAPACITY FOR ‘SELF-CONSTRUCTION’
(metabolism)

POTENTIAL FOR ‘INDEFinite Growth of Complexity’
(‘Darwinian’ evolution)

very complex molecules (DNA, RNA, proteins, sugars, lipids,...)

very complex organization
(‘genetically-instructed’ cellular metabolisms)

[Reprinted (2010): Anthology -- CUP]

LIFE: very complex molecules
very complex organization

LIFE: SYSTEM PROPERTY ! → ORIGINS: SYSTEMIC APPROACH!!
minimal living systems (autonomy + open-ended evolution): ‘TWO/THREE-POLYMER WORLD’ (RNA-protein/DNA-RNA-protein)

‘hereditary autonomous’ systems
‘ONE-POLYMER (RNA) WORLD’

first major bottleneck: ‘proto-bioenergetic’ mechanisms

second major ‘bottleneck’: ‘template-replication’ mechanisms

third major bottleneck: phenotype-genotype decoupling (catalysis /// template activity)

‘transcription’ mechanisms and genetic code

‘basic autonomous’ systems
‘OLIGOMER (peptides) WORLD’

‘PROTO-CELLS’

INCREASE IN MOLECULAR AND ORGANIZATIONAL COMPLEXITY

[ Ruiz-Mirazo et al. (2004) OLEB 34: 323-346 ]
Universal biophysical/biochemical features

SELF-ORGANIZATION & SELF-ASSEMBLY (!)
From molecular to modular cell biology

Leland H. Hartwell, John J. Hopfield, Stanislas Leibler and Andrew W. Murray

Cellular functions, such as signal transmission, are carried out by ‘modules’ made up of many species of interacting molecules. Understanding how modules work has depended on combining phenomenological analysis with molecular studies. General principles that govern the structure and behaviour of modules may be discovered with help from synthetic sciences such as engineering and computer science, from stronger interactions between experiment and theory in cell biology, and from an appreciation of evolutionary constraints.

Timeline | Key events in the application of self-organization concepts in cell biology

- Kant and the self-organized nature of life
- First oscillating chemical reaction
- Turing patterns
- (1972–1977) Biological pattern formation
- (1972–1977) Oscillations in glycolysis
- Exploratory behaviours in cell morphogenesis
- Self-organized microtubule patterns

Self-organization in cell biology: a brief history

Eric Karsenti

NATURE REVIEWS | MOLECULAR CELL BIOLOGY

VOLUME 9 | MARCH 2008 | 255
MAIN ‘BOTTOM-UP’ QUESTION: BEYOND (CHEMICAL) ‘SELF-ORGANIZATION’?....

Chemical ‘self-organization’: coupled autocatalytic cycles

[B-Z reactions + oscillation patterns: control on BC?]
… AND ‘SELF-ASSEMBLY’?
[Ruiz-Mirazo et al. (2008); Moreno & Ruiz-Mirazo (2009) – Biol. & Phil.]
COMPARTMENTS:
(not just ‘hosts’ or ‘containers’ but)
SELECTIVELY PERMEABLE SUPRAMOLECULAR STRUCTURES
THAT DEFINE THE BOUNDARIES OF THE SYSTEM AND ALLOW
ACTIVE CONTROL OF MATTER-ENERGY FLOW THROUGH IT
(TRANSPORT + ENERGY TRANSDUCING MECHANISMS)

‘BOTTOM-UP’ APPROACH

‘co-evolution’

between boundary (scaffolding) and protometabolic reactions
Liposomes from Ionic, Single-Chain Amphiphiles†

William R. Hargreaves* and David W. Deamer†

The First Living Systems: a Bioenergetic Perspective
DAVID W. DEAMER∗
Department of Chemistry and Biochemistry, University of California,
Santa Cruz, California 95064

FIG. 10. Formation of membranous vesicles from mixtures of metocritic amphiphilic compounds. Emulate from plate A of Fig. 9 was dried on a standard glass microscope slide and allowed to interact with a dilute alkaline buffer (10 mM Na2CO3). Under these conditions, the aqueous phase penetrated the dried extract with minutes (top) and vesicle formation ensued over a 30-min period (center). The vesicles contained fluorescent compounds in their membranes, which could be visualized by epifluorescence microscopy with 400-nm blue light for excitation (bottom). If a fluorescent ionic dye such as pyranine was included during the initial hydration, it was captured by the vesicles (not shown).
‘Autopoietic vesicles’ (Pier Luigi Luisi)


THE MINIMAL AUTOPOIETIC SYSTEM

A → S

vägen = \frac{d[S]}{dt}; \quad \text{vdec} = -\frac{d[S]}{dt}

if v_gen = v_dec, \quad \text{homeostasis}

if v_gen > v_dec, \quad \text{growth}

Fig. 4 The minimal autopoietic system. This system is characterized by two competitive reactions, one that builds the component of the boundary, and another one that destroys it. According to the relative value of these two velocity constants, the system can be in homeostasis, or grow, or die

a simple experimental model of chemical homeostasis

Fig. 8 The experimental implementation of the autopoietic model of Fig. 3 with two competitive reactions. Here, one reaction forms new oleate surfactant from the hydrolysis of the anhydride, and one reaction destroys oleate via oxidation of the double bond. Depending upon whether the two velocities are equal or not, different pathways for the systems are obtained—homeostasis (which corresponds to an autopoietic self-maintenance system), growth and self-reproduction, or decay and death (Zepik et al. 2001)
The Emergence of Competition Between Model Protocells

Irene A. Chen,¹² Richard W. Roberts,³ Jack W. Szostak¹*

The transition from independent molecular entities to cellular structures with integrated behaviors was a crucial aspect of the origin of life. We show that simple physical principles can mediate a coordinated interaction between genome and compartment boundary, independent of any genomic functions beyond self-replication. RNA, encapsulated in fatty acid vesicles, exerts an osmotic pressure on the vesicle membrane that drives the uptake of additional membrane components, leading to membrane growth at the expense of relaxed vesicles, which shrink. Thus, more efficient RNA replication could cause faster cell growth, leading to the emergence of Darwinian evolution at the cellular level.

The emergence of cellular behavior. Competition emerges as protocells containing replicating genomes steal membrane from protocells containing inactive molecules.
ENVIRONMENT: a computational platform to stochastically simulate reacting and self-reproducing lipid compartments

Fabio Mavelli\textsuperscript{1,4} and Kepa Ruiz-Mirazo\textsuperscript{2,3}

\textsuperscript{1} Chemistry Department, University of Bari, Italy
\textsuperscript{2} Biophysics Research Unit (CSIC—UPV/EHU), University of The Basque Country, Spain
\textsuperscript{3} Department of Logic and Philosophy of Science, University of The Basque Country, Spain

E-mail: mavelli@chimica.uniba.it and kepa.ruiz-mirazo@ehu.es
Our protocell model: main features/assumptions

1) Realistic diffusion processes (passive transport) across the membrane, considering free flow of water

\[ C_{\text{Total}} = \frac{\sum_i n_i}{N_A V_{\text{Core}}} = \frac{\sum_j n_j}{N_A V_{\text{Env}}} \]

Overall isotonic condition:

\[ \frac{\sqrt[3]{36\pi V^2}}{V_{\text{sphere}}} < S < 2 \frac{\sqrt[3]{36\pi V^2}}{V_{\text{sphere}(V/2)}} = 2 \sqrt[3]{9\pi V^2} \]

\( \Phi = \frac{S}{\frac{3}{36\pi V^2}} \)

\( 1 \leq \Phi \leq \frac{3}{2} \)

2) Conditions for division or an eventual ‘osmotic crisis’

\[ \frac{\sqrt[3]{36\pi V^2}}{V_{\text{sphere}}} = S_{\text{sphere}} < S < 2 \frac{\sqrt[3]{36\pi V^2}}{V_{\text{sphere}(V/2)}} = 2 \sqrt[3]{9\pi V^2} \]

- If: \( S \leq \frac{\sqrt[3]{36\pi V^2}}{V_{\text{sphere}}} \) then: **OSMOTIC BURST!**
- When (or before): \( S = 2 \sqrt[3]{9\pi V^2} \) then: **DIVISION!**

\[ \Phi = S / \frac{3}{36\pi V^2} \]

\( 1 - \varepsilon \leq \Phi \leq (1 + \eta)^{\frac{3}{2}} \)

\( (\varepsilon = \eta = 0.1) \quad 0.9 \leq \Phi \leq 1.386 \)
Minimal cell models

Scheme 0

\[ \begin{align*}
L & \xrightarrow{k_{L\mu}} L_{\mu} \xrightarrow{k_{L}} L \\
B & \xrightarrow{k_{L}} B
\end{align*} \]

Scheme 1

\[ \begin{align*}
X & \xrightarrow{k_{X\mu}} L \xrightarrow{k_{L\mu}} L_{\mu} \\
L & \xrightarrow{k_{L}} B
\end{align*} \]

Scheme 2

\[ \begin{align*}
Z & \xrightarrow{k_{Z\mu}} Z_{\mu} \\
Z & \xrightarrow{k_{Z\mu}} Z \xrightarrow{k_{Z\mu}} Z_{\mu} \\
L & \xrightarrow{k_{L\mu}} L \xrightarrow{k_{L}} L \\
B & \xrightarrow{k_{L}} B
\end{align*} \]

Scheme 3

\[ \begin{align*}
X & \xrightarrow{k_{X\mu}} L \xrightarrow{k_{L\mu}} L_{\mu} \\
L & \xrightarrow{k_{L}} B
\end{align*} \]

Eq. condition:

\[ k_{L\mu} [L]_{aq} S = k_{L} n_{L,mem} \]

\[ \begin{align*}
S & \approx \alpha_{L} n_{L,mem} / 2 \\
[L]_{aq} & = \frac{k_{L}}{k_{L\mu}} \frac{2}{\alpha_{L}} \\
[\mbox{L}]_{eq} & = 0.004 M
\end{align*} \]

\[ k_{L\mu} = 1.0 M^{-1} t^{-1} ; k_{L} = 0.001 t^{-1} \]
‘Empty cell’ dynamics

Spherical membranes with different radius \((R)\) in a pure water solution continuously exchanging lipids \(L\) with the internal core and the external environment. As a consequence, the volume fluctuates around the initial spherical value \(4/3\pi R^3\). In fact, these fluctuations bring small structures \((R \leq 30)\) to collapse due to an osmotic crisis.

In the presence of an osmotic buffer \(B\), the fluctuations of the core volume decrease as the buffer concentration increases and this enlarges size range for cell stability.

Osmotic buffer effect on the core volume fluctuations: \([B]_E = [B]_C = 0.05\text{M}\).

Osmotic buffer effect on the core volume fluctuations: \([B]_E = [B]_C = 0.005\text{M}\).

The average volume fluctuations are reported against buffer concentration.
Reproducing real experimental data: swollen vs. deflated protocell competition dynamics


<table>
<thead>
<tr>
<th>Parameters</th>
<th>Oleic Acid</th>
<th>POPC</th>
</tr>
</thead>
<tbody>
<tr>
<td>$k_{in}$</td>
<td>$7.6 \times 10^3 s^{-1} M^{-1} nm^{-2}$</td>
<td>$7.6 \times 10^3 s^{-1} M^{-1} nm^{-2}$</td>
</tr>
<tr>
<td>$k_{out}$</td>
<td>$7.6 \times 10^2 s^{-1}$</td>
<td>$7.6 \times 10^{-7} s^{-1}$</td>
</tr>
<tr>
<td>$[L]_{Eq} (\phi=1)$</td>
<td>$6.667 \times 10^{-5} M$</td>
<td>$2.857 \times 10^{-10} M$</td>
</tr>
<tr>
<td>$\alpha$</td>
<td>$0.3 \text{ nm}^2$</td>
<td>$0.7 \text{ nm}^2$</td>
</tr>
<tr>
<td>$\nu$</td>
<td>$0.6 \text{ nm}^3$</td>
<td>$1.3 \text{ nm}^3$</td>
</tr>
<tr>
<td>$\varepsilon$</td>
<td>$0.21$</td>
<td>$0.59$</td>
</tr>
</tbody>
</table>

[Chen et al. (2004): Science 305]
A ‘proliferating microsphere’?

[Ganti T. 1975; 2002]


The permeability coefficient to waste results to be a fundamental parameter to guarantee the stability of the cell.

A critical size was found to overcome an eventual osmotic crisis. As much bigger is the size as higher are the stability and growth rate.
Two-lipid membranes: FROM ‘SELF-ASSEMBLY’ TO ‘SELF-PRODUCTION’

\[ k_i = 10, k_w = 0.1, \text{for } i = \{1-6\}; \quad k_{xw} = 7.6 \times 10^{-13} \text{ s}^{-1} \text{M}^{-1} \text{dm}^2; \quad k_x = 4.56 \text{ s}^{-1}; \quad k_{y} = 7.6 \times 10^{-2} \text{ s}^{-1} \]

\[ D_x = D_y = 4.62 \times 10^4 \text{ dm}^2 \text{s}^{-1} \text{mole}^{-1}; \quad D_{xw} = 1.93 \times 10^4 \text{ dm}^2 \text{s}^{-1} \text{mole}^{-1}; \quad D_{yw} = 5.46 \times 10^5 \text{ dm}^2 \text{s}^{-1} \text{mole}^{-1} \]

\[ [X]_{\text{env}} = [Y]_{\text{env}} = 0.001 \text{ M (constant)}; \quad [W]_{\text{env}} = 0 \text{ (constant)} \]

\[ I_{\text{core}} = I_{\text{env}} = 4 \text{ mM}; \quad [A_1]_{\text{core}} = 0.002 \text{ M}; \quad [B] = 0.2 \text{ M (other initial concentrations set to zero)} \]

[Permeability vs. X(flipflip)]

Two-lipid membranes: FROM ‘SELF-ASSEMBLY’ TO ‘SELF-PRODUCTION’

\[ k_i = 10, k_e = 0.1, \text{ for } i = \{1-6\}; k_{A_i} = k_{A_{\text{mix}}} = 7.6 \times 10^{13} \text{ s}^{-1} \text{ M}^{-1} \text{ dm}^2 \text{ s}^{-1}; k_i = 4.56 \text{ s}^{-1}; k_e = 7.6 \times 10^{-2} \text{ s}^{-1} \]
\[ D_{A_i} = D_{A_{\text{mix}}} = 4.62 \times 10^{-4} \text{ dm}^2 \text{ s}^{-1} \text{ mole}^{-1}; D_{A_{\text{mix}}} = 1.93 \times 10^{-5} \text{ dm}^2 \text{ s}^{-1} \text{ mole}^{-1}; D_{A_{\text{mix}}} = 5.40 \times 10^{-7} \text{ dm}^2 \text{ s}^{-1} \text{ mole}^{-1} \]
\[ [X]_{\text{env}} = [Y]_{\text{env}} = 0.001 \text{ M (constant); } [W]_{\text{env}} = 0 \text{ (constant)} \]
\[ [I]_{\text{env}} = [I]_{\text{core}} = 4 \text{ mM; } [A_i]_{\text{core}} = 0.002 \text{ M; } [B] = 0.2 \text{ M (other initial concentrations set to zero)} \]

Physical effects underlying the transition from primitive to modern cell membranes

Itay Budin\textsuperscript{a,b} and Jack W. Szostak\textsuperscript{a,b,1}

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PNAS | March 29, 2011 | vol. 108 | no. 13 | 5249–5254

Fig. 4. Phospholipids inhibit solute permeation through fatty-acid-based membranes. (A) Permeability of C10 membranes (4:1:1 DA:DOH:GMD) to ribose as a function of DDPA content as measured by a stopped-flow relaxation assay. (B) Leakage of encapsulated ImpD from C10 vesicles as measured by scintillation counting of dialysis buffer aliquots. Membrane compositions: \large{□} 4:1:1 DA:DOH:GMD; \large{▲} 4:1:1 DA:DOH:GMD with 25 mol % DDPA; and \large{★}, DDPA.

Fig. 5. Schematic for membrane-driven cellular evolution. The gradual transition from highly permeable primitive membranes (Left) to phospholipid membranes (Right) is driven by the selective growth advantage provided by increasing phospholipid content in the membrane. In turn, this transition in membrane composition imposes a selective pressure for the emergence of internalized metabolism to counter the reduced permeability of diacyl lipid membranes.

Fig. 1. Phospholipids drive competition between fatty acid vesicles. (A and B) Competition between vesicles was monitored by a FRET-based real-time surface area assay. Growth of FRET-labeled 90:10 oleate:DOPA vesicles (A) and shrinkage of FRET-dye labeled oleate vesicles (B) when mixed 1:1 with buffer (black), unlabeled oleate vesicles (green), or unlabeled 90:10 oleate:DOPA vesicles (blue). (C and D) The dependence of vesicle growth or shrinkage on vesicle stoichiometry. Final growth after equilibrium of FRET-labeled 90:10 oleate:DOPA vesicles (C) and shrinkage of FRET-labeled oleate vesicles (D) when mixed with varying amounts of unlabeled oleate (●) or unlabeled 90:10 oleate:DOPA (▲) vesicles. Error bars indicate SEM ($N = 3$).
Template-directed synthesis of a genetic polymer in a model protocell

Sheref S. Mansy¹, Jason P. Schrum¹, Mathangi Krishnamurthy¹, Sylvia Tobé¹, Douglas A. Treco¹ & Jack W. Szostak¹

Figure 1 | Conceptual model of a heterotrophic protocell. Growth of the protocell membrane results from the incorporation of environmentally supplied amphiphiles, whereas division may be driven by intrinsic or extrinsic physical forces. Externally supplied activated nucleotides permeate across the protocell membrane and act as substrates for the non-enzymatic copying of internal templates. Complete template replication followed by random segregation of the replicated genetic material leads to the formation of daughter protocells.
Table 1 Summary of experiments related to gas-phase amino acid syntheses under simulated prebiotic earth conditions.

<table>
<thead>
<tr>
<th>Author</th>
<th>Reactants</th>
<th>Energy source</th>
<th>Results reported</th>
<th>*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Miller [27]</td>
<td>CH$_4$, NH$_3$, H$_2$O, H$_2$</td>
<td>Electric discharges</td>
<td>Simple amino acids, organic compounds</td>
<td>–</td>
</tr>
<tr>
<td>Garrison et al. [33]</td>
<td>CO$_2$, H$_2$O</td>
<td>40 MeV helium ions</td>
<td>Formic acid, formaldehyde</td>
<td>(+)</td>
</tr>
<tr>
<td>Abelson [34]</td>
<td>CO, CO$_2$, N$_2$, NH$_3$, H$_2$, H$_2$O</td>
<td>Electric discharges</td>
<td>Simple amino acids, HCN</td>
<td>–</td>
</tr>
<tr>
<td>Bar-Nun et al. [35]</td>
<td>CH$_4$, NH$_3$, H$_2$O</td>
<td>Shock wave</td>
<td>Simple amino acids</td>
<td>–</td>
</tr>
<tr>
<td>Harada, Fox [36]</td>
<td>CH$_4$, NH$_3$, H$_2$O</td>
<td>Thermal energy (900–1200 °C)</td>
<td>14 proteinogenic amino acids</td>
<td>(+)</td>
</tr>
<tr>
<td>Lawless, Boynton [37]</td>
<td>CH$_4$, NH$_3$, H$_2$O</td>
<td>Thermal energy</td>
<td>Glycine, alanine, aspartic acid, β-alanine, N-methyl-β-alanine, β-amino-n-butyric acid</td>
<td>–</td>
</tr>
<tr>
<td>Groth, Weyssenhoff [38]</td>
<td>CH$_4$, NH$_3$, H$_2$O</td>
<td>Ultraviolet light (1470 and 1294 Å)</td>
<td>Simple amino acids (low yields)</td>
<td>–</td>
</tr>
<tr>
<td>Sagan, Khare [39]</td>
<td>CH$_4$, C$_2$H$_6$, NH$_3$, H$_2$O, H$_2$S</td>
<td>Ultraviolet light (&gt;2000 Å)</td>
<td>Simple amino acids (low yields)</td>
<td>–</td>
</tr>
<tr>
<td>Yoshino, Haratsu, Anders [40]</td>
<td>H$_2$, CO, NH$_3$, montmorillonite</td>
<td>Temperature of 700 °C</td>
<td>Glycine, alanine, glutamic acid, aspartic acid, histidine, lysine, arginine</td>
<td>–</td>
</tr>
<tr>
<td>Kobayashi et al. [41]</td>
<td>CO, N$_2$, H$_2$O</td>
<td>Proton irradiation</td>
<td>Various amino acids</td>
<td>+</td>
</tr>
<tr>
<td>Palm, Calvin [42]</td>
<td>H$_2$, CH$_4$, NH$_3$, H$_2$O</td>
<td>Electron irradiation</td>
<td>Glycine, alanine, aspartic acid</td>
<td>–</td>
</tr>
<tr>
<td>Miyakawa, Kobayashi, Sawaoka [43]</td>
<td>CO, N$_2$, H$_2$, H$_2$O</td>
<td>High-temperature plasma</td>
<td>Glycine, alanine, aspartic acid</td>
<td>(+)</td>
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<tr>
<td>Kobayashi et al. [44]</td>
<td>CO, CO$_2$, N$_2$, H$_2$O</td>
<td>Proton irradiation</td>
<td>Glycine, alanine, aspartic acid, serine, threonine, glutamic acid</td>
<td>+</td>
</tr>
<tr>
<td>Plankensteiner, Reiner, Schanz, Rode [31,32]</td>
<td>CO$_2$, N$_2$, H$_2$O</td>
<td>Electric discharges</td>
<td>Glycine, alanine, valine, serine, proline, lysine, histidine</td>
<td>+</td>
</tr>
</tbody>
</table>

*The probability to occur on prebiotic earth according to recent geochemical data: –, little or no probability; (+), possible under special circumstances; +, possible.
(Pre-biopolymer) scenario with:

**SELF-ASSEMBLING VESICLES**
made of fatty acids, amphiphiles/surfactants, alcohols, mixtures,...
evidence from: (a) external sources [Deamer 1986, 1997; Dworkin et al. 2001]
   (b) abiotic (Fischer-Tropsch) synthesis [Nooner et al. 1976; Allen & Ponnampерuma 1967; Rushdi & Simoneit 2001]

**SHORT PEPTIDE CHAINS** (rudimentary channels/carriers and catalysts)
made of: Ala, Gly, Asp, Glu, Ser, Val…
evidence from: (a) external sources [Pizzarello et al. 2006; Bernstein et al. 2002]
   (b) abiotic (Strecker, SIPF,… ) synthesis [Miller 1953; Rode 1999]

**VARIOUS ‘COENZYME-LIKE’ COMPOUNDS** (e- carriers, pigments...)
**PAHs:** PHOTOCHEMICALLY ACTIVE and MEMBRANE STABILIZING!
**PRIMITIVE ENERGY TRANSDUCTION MECHANISMS** ?
   (‘chemical and chemiosmotic’ -- energy currency precursors)
Why postpone the appearance of compartments when they seem to be pivotal for the material-energetic implementation of a complex reaction system?? (+later on: only makes integration problems worse!)
Modelling minimal self-(re-)producing ‘lipid-peptide’ cells (+ feedback mechanism!)

Feedback mechanism: ‘self-regulation’ of waste release/transport
Chiang et al. 2003
α-Helical Hydrophobic Polypeptides Form Proton-Selective Channels in Lipid Bilayers

A. E. Oliver and D. W. Deamer
Section of Molecular and Cellular Biology, University of California-Davis, Davis, California 95616 USA

ABSTRACT Proton translocation is important in membrane-mediated processes such as ATP-dependent proton pumps, ATP synthesis, bacteriorhodopsin, and cytochrome oxidase function. The fundamental mechanism, however, is poorly understood. To test the theoretical possibility that bundles of hydrophobic α-helices could provide a low energy pathway for ion translocation through the lipid bilayer, polyamino acids were incorporated into extruded liposomes and planar lipid membranes, and proton translocation was measured. Liposomes with incorporated long-chain poly-L-alanine or poly-L-leucine were found to have proton permeability coefficients 5 to 7 times greater than control liposomes, whereas short-chain polyamino acids had relatively little effect. Potassium permeability was not increased markedly by any of the polyamino acids tested. Analytical thin layer chro-

FIGURE 13 Computer-generated α-helical aggregates. Polyleucine α-helices (20 residues) were produced with MacMddad software (Molecular Applications Group, Stanford University, Stanford CA). The backbone structure is shown from above in 13 A, and in side view in 13 B. CPK space-filling versions of the top view were then fitted to show the 3a structure (13 C), described by Furois-Corbin and Pullman (1986), which excludes water, and the 4a structure (13 D), which may include a chain of water molecules. Certain pentameric aggregates (not shown) have ample room to provide an aqueous channel. The bundles of α-helices shown here are for illustration only. No attempt was made to find energy-optimized structures.
Question: ‘RELIABLE REPRODUCTION’ IN PROTOCELLS WITHOUT TEMPLATES OR A COMPLEX METABOLISM ???
General condition for the steady state cycle

\[
\frac{1}{C_T} \left\{ v_R + \frac{S}{N_A V} \sum_i \phi_i \left( [X_i]_{\text{Ex}} - [X_i] \right) \right\} = \frac{\alpha_L V}{2 S} r_L N_A
\]

- \(C_T\) = overall internal concentration of molecules
- \([X_i]\) = internal concentration of the \(i\)-th species
- \([X_i]_{\text{Ex}}\) = external concentration of the \(i\)-th species
- \(\phi_i\) = permeability of the \(i\)-th species
- \(V\) = vesicle volume
- \(S\) = vesicle surface
- \(\alpha_L\) = surface area of a lipid molecule (0.7nm\(^2\))
- \(N_A\) = Avogadro’s Number
- \(r_L\) = metabolic rate of lipid production
- \(v_R\) = metabolic rate of overall concentration change

\(\gamma = 1\) \(\Rightarrow\) STATIONARY REPRODUCTION

\(\gamma < 3/2\)
\(\gamma = 3/2\)
\(\gamma > 3/2\)

\(\gamma = \left( \frac{1}{V_g} \frac{dV}{dt} \right) \left/ \left( \frac{1}{S_g} \frac{dS}{dt} \right) \right. = \frac{S_g}{V_g} \frac{dV}{ds}\)

- \(V_0 = 4\pi \rho_0^3 / 3\)

Stochastic simulations outcomes

MAIN ASSUMPTIONS:
- \(\phi_P \gg 1 \Rightarrow ([P]_{\text{Ex}} - [P]) \approx 0\)
- instantaneous flux of water across the membrane
- fast uptake of lipids by the bilayer

APPROXIMATIONS: e.g.,

\[
\frac{r_L}{C_T} = \frac{\alpha_L V}{2 S} r_L N_A \quad \Rightarrow \quad \rho_0 = \frac{6}{\alpha_L C^T N_A}
\]

FINAL REMARKS

NEED TO DEVELOP

PROTOCELL CHEMISTRY RESEARCH (in silico + in vitro)
TO GAIN BETTER UNDERSTANDING OF:

• ORIGINS OF INCREASINGLY COMPLEX
  PROTO-METABOLIC NETWORKS

• ROLE OF THE MEMBRANE AS A SUPRAMOLECULAR DYNAMIC
  STRUCTURE THAT CAN HAVE VARIOUS FUNCTIONS:
  AVOID DIFFUSION, CONTROL ON MATTER-ENERGY FLOW
  THROUGH THE SYSTEM, CATALYTIC EFFECTS, ...

• ‘PRE-DARWINIAN’ EVOLUTIONARY DYNAMICS:
  BEFORE HIGHLY RELIABLE (BUT SOPHISTICATED) MECHANISMS
  OF REPRODUCTION AND HEREDITY WERE DEVELOPED
minimal living systems (autonomy + open-ended evolution):
‘TWO/THREE-POLYMER WORLD’
(RNA-protein/DNA-RNA-protein)

third major bottleneck: phenotype-genotype decoupling
(catalysis /// template activity)
‘translation’ mechanisms and genetic code

second major ‘bottleneck’: ‘template-replication’ mechanisms

first major bottleneck: ‘proto-bioenergetic’ mechanisms

‘hereditary autonomous’ systems
‘ONE-POLYMER (RNA) WORLD’

‘basic autonomous’ systems
‘OLIGOMER (peptides) WORLD’

function

increase in molecular and organizational complexity

information

origins of life

Sustained life is a property of an ecological system rather than a single organism or species. [Morowitz 1992]

(...) Traditional biology has tended to concentrate attention on individual organisms rather than on the biological continuum. The origin of life is thus looked for as a unique event in which an organism arises from the surrounding milieu. A more ecologically balanced point of view would examine the proto-ecological cycles and subsequent chemical systems that must have developed and flourished while objects resembling organisms appeared.
Thank you!
Fig. 1. Main pathways of energy transduction in living cells. (A) Respiratory and photosynthetic routes that lead to ATP synthesis. (B) ATP is synthesized by glycolysis and other metabolic pathways. (C) ATP is utilized for chemical and mechanical work. 

[References]

[Lipmann 1941]
[Mitchel 1961]
[Harold 1986]

[Skulachev 1992]
WE NEED TO EXPLORE 'NEW (rather messy) CHEMISTRIES':
reactions + diffusion + transport across non-aqueous phases
(beyond 'suspension/surface chemistries' !)

Epstein & Vanag (2005):
'BZ-AOT' system
reaction-diffusion in microemulsions

Hanczyc & Ikegami (2008):
chemistry + self-assembly + convection
(primitive type of chemotaxis?)

[H_2SO_4][NaBrO_3]/[MA], M

Droplet fraction, \( \phi_d \)

Homogeneous Steady State

- Clusters
- Standing waves
- Packet waves
- Oscillation
- Turing structures

Accelerating Waves

- Anti-Spirals
- Dash Waves
- Chaos
- Segmented Circular Waves
- Front of Bubbles
- Segmented Spirals

pH = 10

pH = 11
Definition: self-organisation is a dynamic phenomenon in which a large number of individual units (molecules, cells, multicellular organisms) spontaneously generate a global, irreducible co-relation that brings and holds them together: i.e., a collective pattern of order or behaviour that involves all of those interacting units and cannot be explained just from their individual properties.

'SELF-ORGANISATION'
(Ruiz-Mirazo 2011, Essay for the Systems Biology Encyclopedia)
**Definition**: a protocell is any experimental or theoretical model that involves a self-assembled compartment (typically a supramolecular structure, like a lipid vesicle) linked to chemical processes taking place around or within it, aimed at explaining how more complex biological cells or alternative forms of cellular organization may come about.

‘PROTOCELL’
A MORE COMPLETE PICTURE

AUTONOMY ⇔ FUNCTION (and AGENCY)

Towards a theory autonomy (basic biological organization) well rooted in physics-chemistry: in particular, developing a naturalized concept of function that includes material-thermodynamic aspects (work-constraint cycle, endo-exergonic couplings!)

[Kauffman 2000, 2003]

OPEN-ENDED EVOLUTION ⇔ INFORMATION

Naturalization of the ‘Shannon & Weaver’ (syntactic) conception of information by embedding it in a new theoretical framework that takes into account organizational (semantic) aspects