Mihail Barboiu : From Molecular Machines to Biomimetic Artificial Membranes and Dynamic Constitutional Materials

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As an introduction, one needs to stress that my presentations of Roumanian scientists whom I know personally and who deserve to be better known by chemists in Romania, describe individuals who are actively involved at present in research, so that their achievements are to be preceded by the information: *so far – till now*.

Mihail Barboiu (Mihai for his friends), who is now a Professor and CNRS Research Director at the European Membrane Institute, at the University of Montpellier in France, can look back at his numerous spectacular achievements in organic and supramolecular chemistry despite his relatively young age (49 years in 2017).

He graduated as chemical engineer from the University Politehnica of Bucharest (UPB) in 1993. As a student, in collaboration with Prof. Dr. Claudiu Supuran, he had already started doing research work (with Prof. Constantin Luca, his very first scientific mentor and friend, Head of the Chair of Analytical Chemistry [1] and with me on pyrylium salt chemistry [2]), and he continued this collaboration after graduation when he worked as Assistant Professor for the Analytical Chemistry Department at UPB and collaborated with the Center for Membranes in Bucharest led by Dr. Georgeta Popescu. With membranes being used industrially for water microfiltration, the Center was fairly safe from political pressure.

Thus the very young Mihai became an independent research career, coordinating the first research group and laboratory working on *Supramolecular chemistry and membranes* [3]. This group made good progress on all the projects, mostly centered around biomimetic membranes, for instance those involved in facilitating transport *via* non-covalent interactions of L-amino-acids (fig. 1). In most of his papers during the time he was in Romania, he was the first author since he was the driving force for the research [4-7].

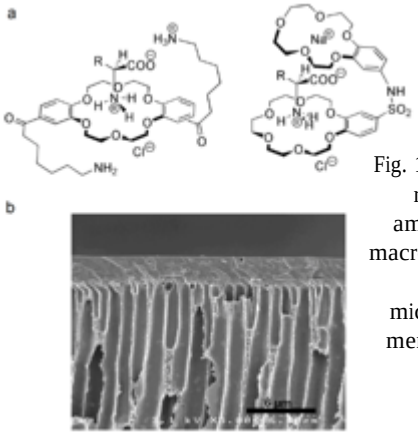

Fig. 1. a) Multiple molecular recognition of the L aminoacids by polytopic macrocyclic receptors and b) Cross section SEM micrograph of the hybrid membrane containing the receptor [4,7]

This Center for Membranes in Bucharest had a collaboration agreement with the corresponding Laboratoire des Matériaux et Procédés Membranaires (LMPM) at the University of Montpellier II (UMII) by Prof. Louis Cot (at present, the European Membrane Institute (EMI) of Montpllier). In 1998 Mihai Barboiu obtained his Ph. D. degree in Chemistry of Materials, jointly from UPB Bucharest and UMII Montpellier (the latter *summa cum laude*, advisors Prof. C. Luca and Prof. C. Guizard). An opportunity of a lifetime opened in 1998 with an INCO-COPERNICUS European Program allowing Mihai to continue his post-doctoral research in Montpellier.

Then in 1999, Mihai joined Professor Jean-Marie Lehn's research group in Strasbourg (along with this, Mihai was offered a position of Assistant professor at College de France in 1999-2001). For his results summarized in his book *Supramolecular Chemistry*, [8] Professor Lehn had been awarded the Chemistry Nobel Prize in 1987, which he shared with Donald J. Cram and Charles D. Pedersen [9].

As a side remark, by analogy with burrs from plants, George de Mestral in Switzerland had invented the hook-and-loop fastener Velcro (from *velours* and *crochet*) [10]. It binds together materials by many weak forces, which open easily individually but are string together. A similar evolutionary adaptation allows geckos to walk on vertical walls. In chemistry, along with the strong covalent, ionic or coordinative bonds, natural evolution relies heavily also on the weaker hydrogen bonds/bridges or the still weaker dipole-dipole forces and London dispersion forces. Thus, hydrogen bonds are responsible for base pairing in cell division (cytosine to guanine, and adenine to uracil in RNA and to thymine in DNA).

Between 2002 and 2006, eight of the 30 Mihai's papers are the result of collaborations with Prof. Jean-Marie Lehn and will be discussed further below. The collaboration continued during the following years, after Mihai had become associate professor at the European Membrane Institute of Montpellier and CNRS Research Group Leader (2001-2008). He became promoted further as professor and senior scientist after 2008. With his Adaptive Supramolecular Nanosystems (ASN) research group at EMI-Montpellier, he has opened new areas in organic chemistry, biochemical and medicinal chemistry.

As an introduction to Mihai's initial results, one must recall that in early 1970s, scientific research in Roumania suffered great losses: many research institutes obliterated (including the Academy's Institutes of Mathematics), and the remaining ones were transferred to various economic departments, so that all research should henceforth be directed towards immediate technological results.

Although Mihai's research fields are interlocked, it will be easier to follow them in distinct sections with italicized titles.

Molecular machines

There are several types of chemically-induced mechanical movements necessary for life. Natural evolution arrived at efficient proteins for these motions, grouped under the name solution dynamics: linear movement (as with actin and myosin involved in muscle contraction and extension), (as with ATP synthase), and helical coiling (helices can extend and contract). Most researchers around the world were working on making artificial supramolecular machines that were able to perform either linear or rotational motions. However, *the most important mechanical motion in biology is the coiling motion of the proteins*, as Professor Lehn, remarked. Mihai's research was centered on the this type of motion and a comprehensive review, summarizing all these works has been recently published [11].

More precisely, a sequence of three covalently-bonded 6-membered heterocycles (pyrimidine-pyridine-pyrimidine) adopting a *transoid-transoid* conformation can have an coiled/helical compact geometry, but in the presence of metal ions (Pb2+, Zn2+, Ag+) or protons, rotation around single bonds so that nitrogen heteroatoms can converge affording complex bonding with a linear (fig. 2a)[12, 13] or double helical (fig. 2b) [14] extended geometries, respectively. Self-assembly of several metal ions and heterocyclic sequences can then amplify the effect up to nanometric lenghts. By controlling the *p*H, one can enforce to alternate the reversible dynamic uncoiling/coiling motions associated with the binding and release of metal ions, eventually orchestrating transitions of helical molecules into grid (fig. 2c) or linear complexes and back again [15, 16]. One of these papers[17] has been dedicated to Prof. Mircea Banciu, Professor for Organic Chemistry Department at UPB, who was a mentor of Mihai during his very first steps in chemical research.

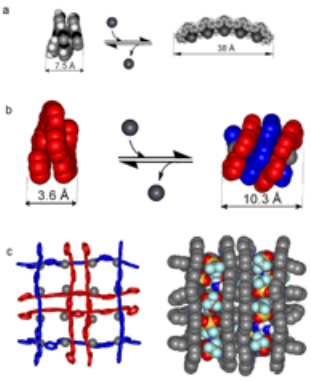

Fig. 2. Reversible extension of the Pyridine-Pyrimidine, py-pym helical strands into a) the linear Pb^{2+} and b) double helical Ag^{+} complexes, respectively. c) X-ray structure of a hexadecanuclear [4x4] Pb^{2+} grid of a py-$(py\text{-}pym)_3$-py-py ligand in stick (left) and in space-filling (right) representation including the triflate anions bound in the cavities of the assembly

Dynamic constitutional materials and membranes.

After this incipient period, Mihai returned to the biomimetic systems research. In 2004 he was awarded by the European Science Foundation and the European Heads of Research Councils with the very prestigious *European Young Investigator Award*, the former European Research Council ERC Grants. Within this context, Mihai' research was mainly dedicated to *Dynamic constitutional chemistry toward functional materials and membranes*. Concerning this field, Mihai is the Editor of a unique successful book on *Dynamic Constitutional Chemistry*. His research has moved toward using dynamic interactive systems for innovative materials and functional devices The main aims have been devoted to make *biomimetic membranes and sensors*, [18 - 20] but have also moved into new fields.

In a broadest sense the self-organization play an important role at different scales in living matter, biological information storage and biomolecule self-organization. The ASN Group at leaded by Mihai, highlighted important accomplishments in the field of biomimetic hybrid membranes and materials. Mihai concentrated his efforts on studies related to constitutional self-organization of nucleobase (fig. 3a), [21-23] peptide (fig. 3b)[24, 25] or *crown-ether* (fig. 3c) [26, 27] dynamic libraries. The objectives were to expand the molecular self-organization toward nanoscale and to express the fittest adaptive selection of highly functional architectures.

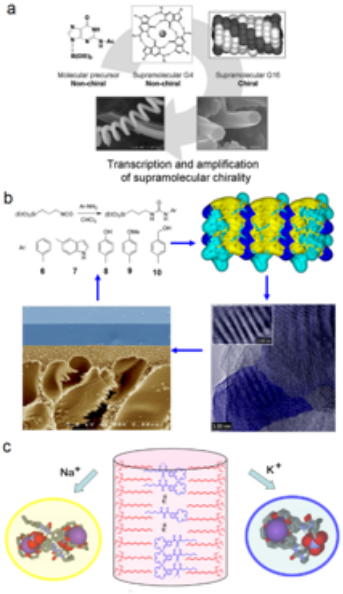

Fig. 3. Dynamic Constitutional Materials. a) The K^{+} cation-templated *G-quadruplex* transcription in solid chiral hybrid materials; b) multiscale nanostructuration of peptoid thin-layer hybrid membranes and c) Constitutional reorganization of mesopore confined macrocyclic ion-channels evolving an adaptive response in the presence of an ionic solute

Important efforts were concentrated on the elaboration of self-organized *ion-channels at the nanoscale* [28] or of the *hybrid constitutional materials* [29-33] with particular emphasis focusing on flexible functionality and adaptation within confined conditions.

The ASN group has also joined as Coordinator, important European RTN Marie Curie Networks: DYNAMIC and then DYNANO, [34] privileging new projects on *Dynamic Constitutional Nanosystems*: they unlock the door to the new world paralleling that of biology. Within this context the *sugar-protein interactions* that mediate *cell-cell recognition* generated a new trend that Mihai' group is following along a fruitful collaboration with industrials and scientists [35-38]. Constitutional dynamic libraries (CDL) have also been used for drug discovery purposes and several papers have been published (Back into the future!) in cooperation with his very old friend Prof. Claudiu Supuran on *Dynamic Constitutional Discovery* of Carbonic Anhydrase Inhibitors and Activators [39-41].

Dynamic Metallosupramolecular Libraries (DMLs) can be generated *via* metal–ligand and reversible covalent ligand bond exchanges. The metal ion coordination process may help to position ligands in close proximity. Reversible molecular exchanges and supramolecular non-covalent interactions may occur synergistically, leading to rearrangements of the coordinating ligands within a restricted interactional space, defined by the coordination geometry around the metal centers. A rather unconventional and as yet relatively uncharted option is multiple coding, offered in the process of simultaneous coordination with mixtures of ligands, producing DMLs of mono- and poly-nuclear components containing distinct metal ions [42-44].

Concurrently, the use of Dynamic Constitutional Frameworks for multivalent biological interactions has attracted a great deal of interest [45]. Within this context, the Dynamic Constutional Chemistry, appeared as one of the most attractive screening method for the rapid access

to active systems designed to mimic natural DNA delivery, [46-48] protein recognition [49] or dynamic biopolymers [50].

Artificial water and ion-channels. Our bodies consist mostly (two thirds) of water which, due to hydrogen bonds, is a liquid under a wider range of temperatures than most other substances. Life, as we know it, could not exist without water. It crosses the bilayer cell membranes at a slow rate that depends on several factors including osmotic pressure. Some cell membranes, e. g. in kidneys and red blood cells, evidence very high diffusion rates because they contain protein pores allowing the passage of water molecules (aquaporins) as well as solvated ion solutions (ionic channels). Aquaporin-1 is a membrane protein that has the shape of an hourglass with a central narrow channel (diameter 2.8 Å) that allows only water molecules, without any solvated ions. For a stretch of 20 Å, a single file of water molecules crosses the cell membrane, but the efficiency of a channel is awesome: hundreds of millions of molecules per second! For the discovery of aquaporins, Peter Agre was one of the two Chemistry Nobel Prize laureates in 2003 [51].

As a side remark. professor Gheorghe Benga from Cluj, titular member of the Roumanian Academy, had published earlier results in this field, but had not obtained purified aquaporin-1. Like Nicolae Paulescu (discoverer of insulin) or Stefan Procopiu (discoverer of magneton) in the previous century, Professor Gheorghe Benga was overlooked by the Nobel Prize Committee.

Mihai's pioneering research for artificial water channels is based on his familiarity with membranes and with supramolecular self-assembly. Since the fresh water global deficiency is already felt by many countries, and since desalination of sea water using reverse osmosis is still expensive, artificial water channels are a *hot topic*. Mihai has published the first papers [52,53] and two reviews on artificial water channels (fig. 4) [54,55] for which he received in 2015 the Royal Society of Chemistry Surface and Interfaces Award [56].

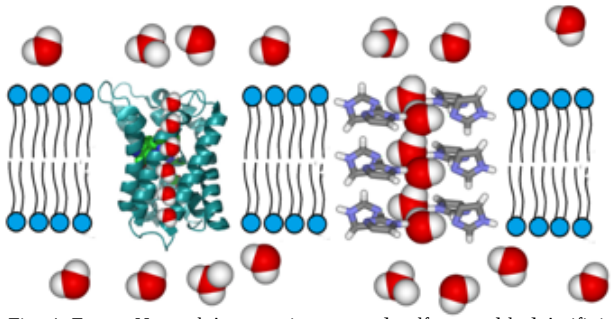

Fig. 4. From Natural Aquaporins toward self-assembled Artificial Water Channel systems. Adapted from reference [55]

So far, they are about a hundred times less efficient than aquaporins. Instead of proteins, the artificial biomimetic membrane water channels are based on simple aromatic and heterocyclic compounds (imidazole, pyridine) that are able to self-aggregate. Moreover, correlations between the artificial channel superstructures and the well-studied aquaporins, might allow designing novel mechanisms that parallel to natural water-transport processes [57]. Conversely, the molecular-scale hydrodynamics of confined water is of crucial relevance for understanding complex functions at the heart of interesting biological scenarios.

Molecular encapsulation is a fascinating domain as the behavior of the related integration of guests are generally different inside capsules compared with the bulk solution. Crossing the solution/capsule barrier, unexpected dynamic phenomena can be observed within a *compartmentalized* chemical space, allowing access to new emergent areas of chemistry and physics under confined conditions. Compartmentalization is also a basic feature of biological processes, as most of the physiological processes occur in cells and depend on selective exchanges of metabolites between the cell and its exterior.

Alkane compression and encapsulation

Lipid-binding proteins encapsulate hydrocarbon chains by using weak hydrophobic and anchoring electrostatic or H-bonding interactions. Dynamic *bridge*, *loop* and *tail* conformations of confined alkanes can be adaptively obtained on guest encapsulation. Spectroscopic NMR studies in aqueous solution have been performed; however these structures have been eluded crystallographic characterization.

Mihai' group solved this problem and recently reported for the first time the exact coiling behaviors of compressed alkanes within crystalline *Pyrene-box* capsules, determined from atomic resolution X-ray diffraction (fig. 5) [58, 59]. The lids of the box are two pyrene-tetrasulfonic acid molecules, amn the side-walls are eight guanidine molecules. The box self-assembles due to the electrical opposite charges, and it encapsulates molecules that happen to be near, influencimg their geometry, as proved by X-ray crystallography and NMR spectra. They revealed zigzag-like conformations of alkanes in the compressed states. H-Bonding be in the neighborhood. interactions between capsule components and van der Waals interactions between alkane guest and the capsule as being the driving force for the assembly of the stable biomimetic capsules in water.

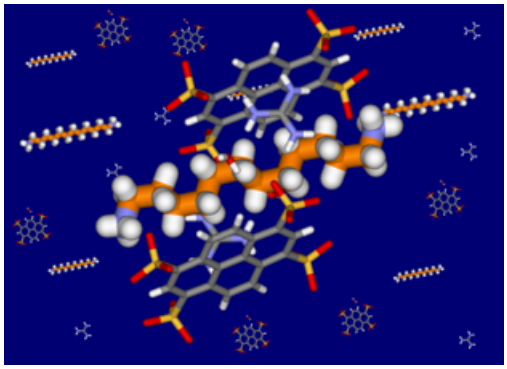

Fig. 5. Compression of confined alkanes: X-ray structure of 1,ù-diammonium guest inside a H-bonded Pyrene box capsule. Adapted from reference [58]

Cyclobutadiene structure

The stabilization of benzenoid hydrocarbons is due to ring-closed electronic conjugation, as offered for instance by an odd number of double bonds in anulene rings such as benzene (CH)6, the prototype of aromatic hydrocarbons. By contrast, a planar hydrocarbon with an even number of double bonds would be destabilized. Mihai provided the most direct proof by X-ray crystallography that 1,3-dimethyl-cyclobbutadiene can exist in an anti-aromatic square-planar geometry, or in a rectangular bent and non-conjugated geometry [60, 61]. In the 1960s synthetic advances (including those by C. D. Nenitzescu [62], had shown that cyclobutadiene was extremely unstable and very reactive.

Mihai and his team confined 4,6-dimethyl-2-pyrone in a guanidinium-sulfonate-calixarene at low temperature (175 K); then UV irradiation caused an isomerization to a bicyclic â–lactone, which was stable enough to have its structure established; further UV irradiation caused splitting,

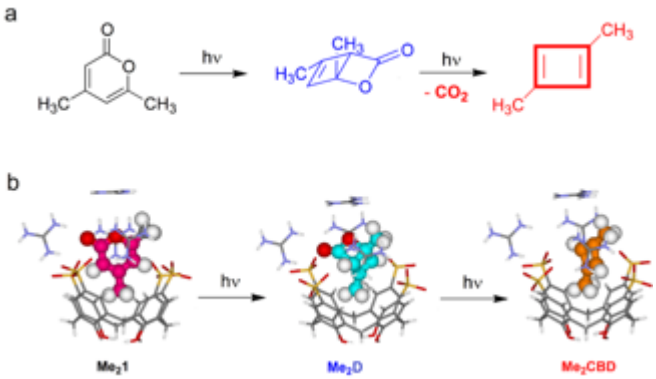

Fig. 6. a) Electrocyclic rearrangement and b) X-ray crystallographic observation of the Me_2CBD formation pathway from photolysis of $Me_2$1 *via* Me_2D. Crystal structure in stick representation of the $G_4C\{Me_2 1\}$ which by UV irradiation transforms progressively into the $G_4C\{Me_2D\}$ and $G_4C\{Me_2CBD\}$ host-guest complexes stabilized under confinement by the Guanidinium-Tetra-*p*-sulfocalix[4]arene, G_4C host matrix. Adapted from reference [62]

affording carbon dioxide and a confined 1,3-dimethyl-cyclobutadiene with two interconverting forms that were characterized by X-ray crystallography: a square-planar geometry (62.3%) and a rectangular-bent geometry (37.3%). Figure 6 displays these steps, omitting the unstable transient intermediates (ketene and zwitterionic cyclobutene carboxylate). In each reaction step, two electron pairs are involved, in agreement with the Woodward Rules for pericyclic reactions: photochemical processes involve even numbers of electron pairs and lead to antarafacial-conrotatory geometries, whereas thermal processes involve odd numbers of electron pairs and lead to suprafacial-disrotatory geometries.

Among the honors obtained by Mihai till now, I would like to mention: Fellow of the Royal Society of Chemistry in 2015; recipient of the Academic Merit Prize- Roumanian Academy, in 2008 and of the Costin D. Nenitescu Medal of Romanian Chemical Society in 2007; decorated in 2016 with National Order for Faithful Service by the President of Romania, Professor Mihai Barboiu, belongs to the pioneers of biomimetic membranes and of constitutional materials for which we can expect in the near future major advances.

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