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# Molecular Interactions – Bringing Chemistry to Life The Workshop Summary

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## Introduction

Molecular interactions, a notion which is familiar to any chemist, many a physicist, and most biologists, probably since the early days of Emil Fischer, has been discussed enthusiastically ever since [1]. But is it worth having a workshop on this topic still today and does it really create any new momentum "bringing it to life"? The answer, being far from simple, I would like to split it into two parts. Firstly, I would like to reflect on the role of molecular interactions in the contributions to the workshop, and secondly, I would like to comment on the efforts taken in the context of the workshop, to bring chemistry to life. The order in which the individual contributions are mentioned is not the order of the workshop programme, but follows my personal view of the topics.

## Workshop Statistics

Under the given heading Bozen 2006 was the meeting place of 36 experts in the field of molecules. More than 70% of them had their roots in chemistry, some 15% in biochemistry, about 10% in biology, and just 5% in physics (Fig. 1).

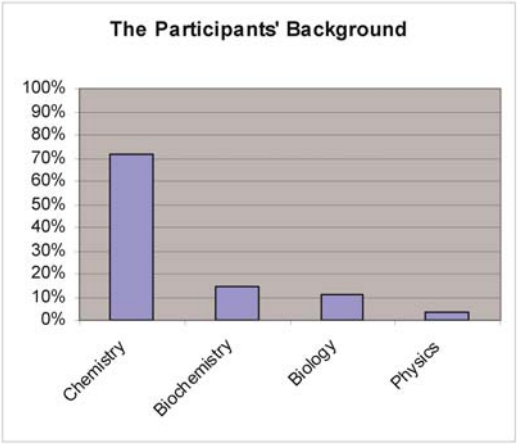


**Figure 1**

From another view point, 47% are employed by universities, 28% by public research institutions, and 25% by industry (Fig. 2). In other words, the majority of participants came from universities and had a background in chemistry.

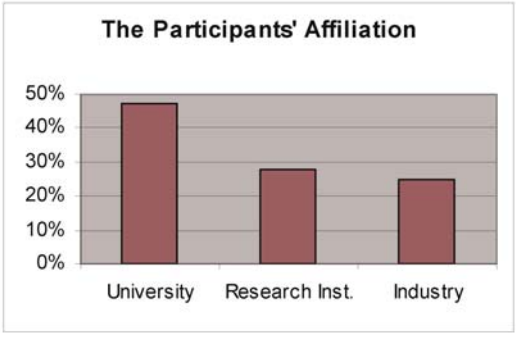


**Figure 2**

For most of the participants the Bozen workshop 2006 was an opportunity to present and discuss new ideas and concepts. The chance was taken by a number of contributions.

## Molecular Interactions

### *Interaction Partners and Scenarios*

On a microscopic scale, it is molecular interaction that forms the basis for the largest part of chemistry, as well as biology. Molecular interaction comes in diverse scenarios. At first glance, there are scattering and reaction, or recognition and binding, using a more biolo-

gical terminology. The interacting partners can be single atoms, small molecules, medium sized molecules, big molecules, molecular ensembles, living cells, or even organisms. From all possible combinations of this list (= 21+1[1] = 22, Fig. 3), 17 have been covered by the talks:

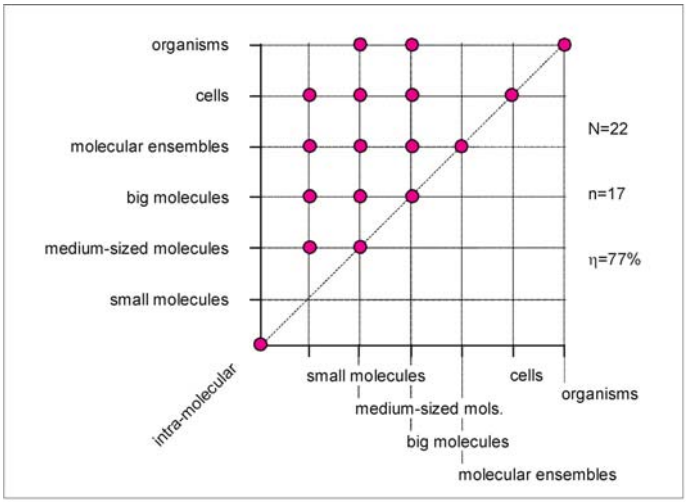


**Figure 3**

To quantify the strength of molecular interaction, the free binding (or dissociation) energy can be used. Figure 4 shows a compilation of typical values. Quantitatively, there is no clear distinction between non-bonding interactions and covalent bonds.

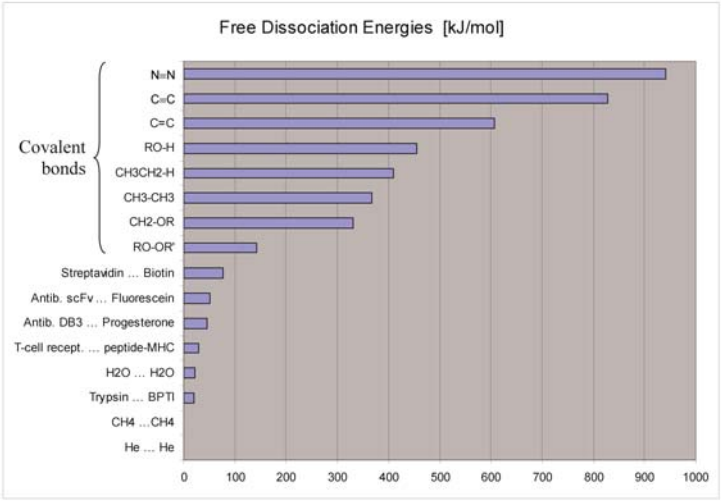


**Figure 4**

[1] All contributions about intramolecular interactions are symbolized by the single spot in the lower left corner.

Real biological systems like living cells deserve some comment. By means of atomic force microscopy it is possible to analyse the interaction of single molecules with individual cells. In particular, one can determine the force necessary to pull a molecule off the surface of a cell [2]. The associated work $W$ is a measure of the height of the barrier that has to be overcome:

$$W = F \times d$$

Here, $F$ is the force applied and $d$ the displacement at which rupture occurs. Unfortunately, the situation for biological systems is rather complicated. In practice, often more than one barrier is encountered and it is typical that barriers change, if an external force is applied [3]. As a consequence, it is very difficult to determine free binding energies. Entropic contributions are complicated and often not fully reproducible. There is some analogy to (α-)relaxation phenomena in semi-crystalline polymers [4].

In addition, the definition of standard conditions with which most people are familiar, from thermodynamics and statistical mechanics is not applicable for biological systems, because these force experiments take place far away from equilibrium.

For the case of cell-cell connection it is known that the interaction is realized by surface receptors and special connector molecules like the cadherins or fibronectin. As a consequence, the actual values of binding energies depend on the surface concentrations of these molecular entities.

***Structures and Interactions***

Nature's capability to compose functional networks based on protein interactions with high reliability, specificity and efficiency clearly demonstrates the potential given by the repertoire of elementary interactions that are possible for molecular structures which are not bound covalently:

- Repulsion of closed electron shells (van-der-Waals repulsion),
- Charge-charge interactions (Coulomb interaction),
- Charge-multipole interactions,
- Multipole-multipole interactions,
- Liquid-crystal-like aromatic interaction (π-π interaction),
- Aromatic multipole-multipole coupling,
- Electronic dispersion (van-der-Waals attraction),
- Vibrational dispersion (oscillator coupling) [5],
- Entropy-related interactions (hydrophobic effect),
- Hydrogen bonding [6].

Needless to say, molecular interactions can never be fully explained in terms of just one or two of these types of elementary interactions.

Numerous crystal structures of proteins involved in networks have helped to uncover and understand the variation of protein-protein interaction scenarios well approved in biological systems throughout the course of evolution. **Tom L. Blundell** showed in his lecture, how these scenarios can be used successfully to design ligands for proteins with therapeutic relevance. In particular targeting of flexible regions of protein surfaces seems to be a promising approach to establish new pharmacological concepts based on oscillator coupling. Even though one has to cope with relatively low binding enthalpies, entropy contributions are able to provide the specificity necessary for lead identification. A new view of target and lead is emerging.

**Oliver Seitz** showed how a carefully designed chemical strategy can be used to trace mutations in DNA in a very efficient manner. A set of specific probes pairing with the DNA target to be analysed can be ligated to form a contiguous double strand if no mismatches (mutations) are encountered. The method has been applied successfully to identify mutations in the RAS gene. Product inhibition after ligation of the probes is prevented by a subsequent rearrangement reaction. The whole cascade can be seen as an example of chemistry based on a strategic combination of interactions moving from a chemical reaction to a chemical system. The method can also be used in combination with PCR and has a potential for predictive diagnostics.

**Joelle N. Pelletier** reported her efforts, to improve the understanding of enzyme-ligand interactions which are of interest in the context of drug resistances. The active site regions of DHFR (involved in tumours resistance against cytostatic drugs) and β-lactamase (key role in bacterial resistance against β-lactam antibiotics) have been mutated systematically by a combinatorial scheme. Even though resistances are a permanent threat for therapeutic concepts and a never-ending challenge for pharmacological research, the situation is not totally hopeless. The chemical space available to nature (here tumours and microbes) is limited by the canonical amino acids and sometimes a finite number of carbohydrates [7]. There is a good chance to explore exhaustively the scope of affinity and specificity of those enzymes, because they are confined to the functions they have been assigned to by evolution. NMR studies and co-crystals with ligands will surely provide deeper insight into the interaction scenarios. However, in many cases of cancer drug resistance, drug resistance is not a local, bilateral phenomenon (one ligand – one target), but rather a systemic issue where transport phenomena are important [8]. In particular for the treatment of cancer, enzymes (kinases) involved in signalling have been targeted for many years and (multi) drug resistance occurs during medication sooner or later. There is a need, hence, to generalize and apply this kind of analyses to other classes of drug targets, e.g., the GPCRs (G-protein coupled receptors) to support also other therapeutical concepts for cancer such as activation of apoptosis and inhibition of angiogenesis [9].

A remarkable interface of biology and chemistry was presented by **Nediljko Budisa**. Non-natural amino acids can be introduced into the process of protein genesis of unicellular organisms resulting in the expression of proteins. A critical aspect of sequence specificity of protein genesis is the substrate specificity of the aminoacyl-tRNA synthetases which link the correct amino acid covalently to the corresponding tRNA adaptors. It is known that at least some of the aminoacyl-tRNA synthetases show a certain substrate tolerance which allows the introduction of non-natural amino acids. Some micro-organisms, so-called auxotrophs, which depend on the uptake of amino acids that they cannot synthesize by themselves, do incorporate 'acceptable' non-natural amino acids, at least if the original encoded amino acid is not available to them. Thus, by adding non-natural amino acids to the nutrition, one can systematically modify expressed proteins. In this way, sites for specific chemical derivatization can be introduced, thereby creating a platform for combined protein genesis and protein synthesis with entirely new perspectives for protein chemistry.

**Marcey L. Waters** took the audience on a tour through the subtle details of molecular recognition. The question she has raised is, can affinity and specificity between biomolecules and synthetic peptide ligands be enhanced systematically at the same time? The most simple model system one can think of is a peptide that folds back on itself to form a hairpin. By NMR and other spectroscopic methods she studied the interaction of a positive charge (Lys) with π-systems (Trp). A competition exists between the coulomb interaction of the ammonium group with the electron density of the π-system of Trp on one hand, and the solvation of the ammonium group together with an interaction of the slightly polarized H-atoms of $C_\varepsilon$ with the electron density of the π-system on the other hand. Unless the ammonium group was totally methylated ($R\text{-}N^{\oplus}(CH_3)_3$), the latter conformation was always found. Analogous findings with systems containing no net charges but carbohydrates instead of Lys show that these combinations of competing interactions based on enthalpy/entropy balance have a substantial impact on the properties of biomolecules. It will be very interesting to see, what other competing combinations will be found. Successful molecular design of drugs has to take care of such constellations.

Structures of possible biological relevance do not necessarily have to consist of biomolecules. **Andreas Hirsch** showed how micelles with characteristic topologies and size can be constructed systematically from non-biogenic amphiphilic molecules. Substituted fullerenes constitute the building blocks for a large variety of stable micelles which may be a signpost for future drug delivery systems.

**Sijbren Otto** presented his work on the design of artificial receptors, generated by a combinatorial strategy. To obtain optimally binding receptors for given ligands, a library of bi-functional building blocks (cyclic peptides) is prepared. In a first step of supramolecular assembly the building blocks aggregate around the ligand molecules. In thermodynamical equilibrium, the most stable products of this recognition step can be found. The next step is the covalent connection of the building blocks with each other by the formation of disulphide bonds from terminal SH-groups of the building blocks. Sijbren Otto has in mind a modular receptor model with an inner shell in direct contact with the ligand and an outer

shell stabilizing the inner shell. This implies some kind of hierarchy of interactions which should result in a multi-step process of ligand binding, controlled by an entropy-driven rearrangement of the receptor. The idea is brilliant, but the realization is very hard to achieve, especially if one is thinking of a self-optimizing scheme. The big challenge in this undertaking comes from its two-dimensional nature. One dimension is the building blocks' affinity to the ligand, a '*conditio sine qua'* the subsequent ligation step is useless. The other dimension is in ligation of the building blocks themselves, which is controlled by reactivity and proper relative orientation of the building blocks. Thermodynamical equilibrium is not necessarily helpful for this second step. There is no guarantee that both dimensions can be optimized at the same time, but the situation is not hopeless. The building blocks have to have a sufficiently large number and suitable choice of (internal) degrees of freedom. Finding the correct mixture is surely a formidable task, but not precluded by the laws of thermodynamics and quantum mechanics. Probably a criterion of selection other than thermodynamical stability has to be applied. Nature's answer to this problem is well known. However, antibodies are much larger than just cyclic peptides and the combinatorial aspect comes with somatic mutations. In other words, nature has a very complex infrastructure available. Nonetheless, this kind of artificial receptors can open up entirely new routes in pharmacology.

A very particular kind of interaction scenario was discussed by **Richard A. Goldstein**. He has studied the evolution of the influenza virus since the 1918 pandemic. The course of this evolution can be traced by a series of transfer events from birds to humans and subsequent outbreak events. A high mutation rate is characteristic of influenza viruses, quickly changing their immunogenic characteristics. In the case of co-infections, influenza viruses can even take up genome fragments from other viruses during replication, resulting in a tremendous speed of evolution. It is easy to imagine, what kind of a challenge the development of vaccines represents. Understanding the systematics of observed changes in the virus strains surely can support preventive healthcare strategies and efficient vaccine development.

Glycosylation of cellular surface proteins is a very important aspect in signal transduction and cellular communication. Interestingly, 75 % of the known cell-surface glycosides can be made of only 36 different carbohydrates. It is, therefore, not surprising that automation of glycoside synthesis is a powerful infrastructure for (glyco-)biological research. **Peter H. Seeberger** presented his solid-phase synthesis robot for all relevant classes of glycoconjugates. Significant improvements in bio-analytics, diagnostics, and vaccine development have been achieved already or will come into reach in the near future. High-throughput strategies, which are well established and successful for nucleotides and peptides, can now be applied to this class of biomolecules, as well. A deeper understanding of interactions in biological systems can be expected.

## Molecules, Signalling and Information

In biology there is a well-known connection between molecular interaction and transfer of (genetic) information. The two dimensions of propagation of genetic information are replication [10] and development (Fig. 5).

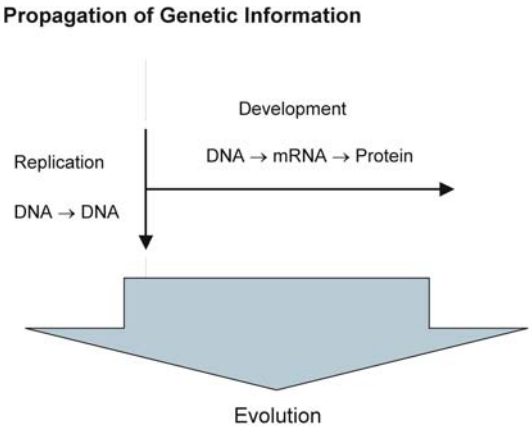


**Figure 5:** Propagation of Genetic Information

In development there is still another mode of transfer, DNA→RNAi, which is known as a means of control of gene expression. Furthermore, both dimensions of information transfer are important for evolution, since selection always is effective on the level of the individuum and hence affected by replication as well as development. It should be noted that the transfer of genetic information is strictly unidirectional. Feedback is a matter of selection.

Of course, there is also propagation of non-genetic information in living organisms:

- Chemotaxis
- Defence
- Repair
- Mating
- Consciousness [11]

**Wilhelm Boland** gave a very detailed insight into the mechanisms and strategies of *Arabidopsis thaliana* in defence against herbivores. The emission of volatile substances to attract enemies of the herbivore is surely an example of plant-insect communication. It does not take too much imagination, to realize that here a focus for future research strategies in crop sciences and veterinary pharmacology is given.

But is interaction of molecules already a form of information transfer and communication? At this point some clarification of the terminology seems appropriate [12]. At the lowest level there are signals. The information is carried by structures and correlations in signal sequences, in other words by semantics, which has to be defined before information can be 'understood'. The lowest level of understanding is the distinction from noise. In the entropy-view of Shannon [13], e. g., information results from the difference between expected and actual signals. Clearly, interaction of individual molecules is a transfer of signals. Information, however, forms at a higher level. Invoking the sender/receiver paradigm, one could say that information originating from a sender is a set of signals which causes any change or response in the receiver [14]. The transfer of information from chromosome to ribosome surely matches this criterion.

The transfer of genetic information is not restricted to the replication or developmental route. Horizontal gene transfer has played an important role ever since life appeared on earth. Microbes and viruses are specialists in transgressing species boundaries and horizontal gene transfer was probably an important mechanism in the beginning of evolution [15]. Today, horizontal gene transfer is about to be used as a therapeutic concept to correct genetic disorders or deficiencies. Usually the method of viral infection is used to transfer the desired genetic information built into a viral genome [16]. The whole process is of paramount complexity. A huge number of possible side reactions has to be taken care of. Since the target is always a living cell, it is hard to imagine that only a handful of interactions are responsible for success or failure. A good example was given by **Laurent M. Humeau**. He presented a gene-therapeutic approach to the treatment of HIV using a lentiviral vector. The concept is based on generating modified T-cells that inhibit HIV replication and has reached clinical phase II. The combination, however, of human intervention and biological mechanisms still presents a challenge [17].

## Models and Representations

Models have always been important tools in understanding molecules and their interactions. Besides visualization, models can be used in the context of perturbation theory, especially in combination with simulation techniques. Applying perturbations to model systems and analysing the model's response is a method used in physics, chemistry, and systems biology, complimenting the vast collections of data available today.

Having its roots in pharmacokinetics, models for biological systems, composed of differential equations are a basis to study phenomena of coupling and regulation in biology. **Athel Cornish-Bowden** showed how to formulate a kinetic system which is capable of self-repair. He raised the question “What does it mean to be alive?” to convince the audience that life in the sense of biology still is far from being representable by models. Even though modelling can offer deeper insights into individual aspects of life, no system can yet be formulated that accounts for all of life's primary characteristics: self-construction, metabolism, self-repair, replication, and evolution [18].

With respect to structure/activity relationships of molecules there is a long tradition in chemistry and physics of model building and simulation [19]. It has been extended on the one hand to polymers of material science and on the other hand to polymers of molecular biology, to address the issue of predicting self-organization (e. g., protein folding) and structure.

A crucial aspect of molecular models is the representation of the system to be studied. The level of complexity defines the number of parameters necessary to map the situation and phenomena of interest. This can be done in a qualitative as well as a quantitative way.

**Stephen J. Haggarty** proposed a graph-theoretical scheme to predict *in-vivo* interactions of molecules based on cluster analysis of annotated interaction data from chemical genetics. Descriptor representations of molecules that interact with synaptic proteins are used to relate a network of local interactions to clusters in biological protein space. Assigning new molecules to identified clusters of interactions and activity should offer the possibility to predict their effect in living organisms.

For the sake of validation, models have been assigned potentials and parameters that are the basis for rating their usefulness in analogy to the (thermodynamical) stability of real systems. Energy minimization and relaxation are first steps of making models similar to their real reference systems in terms of stability. In the traditional hierarchy of methods to estimate energies of atomic and molecular systems, one starts from a quantum mechanical model of nuclei and electrons described by the solutions (wave-functions) of a Schrödinger equation. Looking at increasingly complex systems, one is forced to apply more and more stringent approximations to eventually arrive at a representation with a manageable number of parameters. Careful use of approximations helps to create models, capable of mapping the system under investigation with a level of fidelity that still allows making useful predictions.

**Martin J. Field** showed his results from studying an enzyme reaction which is a key step in replication of *Plasmodium falciparum*. In the course of the reaction a purin base is transferred from nucleic acid of the host to the nucleic acid of the pathogen. The study is aimed at the specificity profile of this reaction to get ideas for new compounds active against malaria. A combination of quantum chemistry for the aspects of reactivity, and molecular mechanics for the aspects of complementarity was used. This kind of hybrid models is surely useful to reduce the model parameterization and complexity to a reasonable size. The critical aspects, however, are related to the influence of nuclear motion. The majority of force field parameters for the mechanical part of the model are derived from either crystal structures or high-precision quantum chemical calculations. In any case, they are adapted to static situations. Elementary steps of chemical reactions, however, are determined by both, the quantum mechanical character of electrons and the dynamical properties of the nuclear framework. This latter phenomenon is actually the domain of scattering theory, which even standard quantum chemistry methods based on the stationary Schrödinger equation in combination with the Born-Oppenheimer approximation cannot describe properly. To obtain results which are mainly free of methodological artefacts, one

has to relax the relevant part of the system along the reaction pathway step by step. At best, this should be done by quantum chemical methods equipped with a gradient optimizer. Another strategy, employed by Martin J. Field, to stay on the safe side, is to confine on trends, comparing similar systems in equivalent states. Coupling the numerical results to experimental data can then be the key to useful predictions.

The contributions of **Jonathan W. Essex** and **Mark S.P. Sansom** have shown that concepts of scaling theory have found their way successfully from pure polymer physics to the assessment of realistic biological questions like passive diffusion through phospholipid membranes and the dynamics of membrane proteins, including aspects of self-organization and folding. It is not easy, however, to stay clear of model-specific concomitants. The approximations' range of validity has to be cross-checked in every case to retain maximum predictivity. A possible remedy could be to switch between different levels of graininess [20]. Atomistic details are needed to find optimal local interactions coupled to high frequency periodic modes, whereas coarse grained representations can account for slower, diffusion-like motions responsible for wider displacements. A lot of knowledge about mechanisms of drug resistance, problems of bio-availability, and mechanism of pathogen attack can be expected from such kinds of simulations, because dynamical phenomena are very likely to play major roles.

Molecules have a fractal surface which is the platform for numerous phenomena based on local interactions. In his lecture, **Tim Clark** took a closer look at the role of molecular surfaces in biological communication. In particular, he is seeking the relationships between surface properties of ligands and their functions in biological systems. The starting point is a classification of the ligands in terms of surface features. As the basis for representation of molecular surfaces he takes linear combinations of spherical harmonics $Y^m_l$. The advantage of such an expansion, either one-centre or multi-centre, is the completeness of the basis. Furthermore, geometrical interpretation of the leading terms provides a very pictorial representation of the relationships and can easily be translated back into structural features due to the directionality of spherical harmonics. This is a great improvement with respect to the classical triangulation-based methods of surface representation, which, with the exception of mere surface area, are much harder to interpret. It will be interesting to see what kind of structure-activity relations can be extracted on the basis of such a kind of representation.

Another intramolecular interaction scenario was analysed by **Jonathan P. Clayden**. He has synthesized oligomeric aromatic amides that arrange the relative orientation of the amide moieties by dipole-dipole interaction. Interestingly, the amide orientation can be propagated along the sterically stiffened backbone of the molecules through at least 22 bonds. There is evidence that the conformational state can even be induced to neighbouring molecules. Again, this is an example of signal propagation that can be used in principle for the purpose of information transfer and storage, including switchable dyes. Apart from nanotechnological applications, one can think of quite a different type of use in synthetic chemistry. The phenomenon described could be used to create tunable catalysts that can be modified during the course of a catalysed synthetic process by either external control or feedback.

**Gisbert Schneider** presented his efforts in the search for new active compounds derived from natural products, a strategy, which has experienced renewed interest [21] only recently. His particular strategy is to change the backbone architecture for given reference compounds, which results in a redefinition of the lead structure concept. A 2- or 3-point representation of the pharmacophore serves as a search template against a structure database of active compounds. Similarity is measured based on a virtual ligand scheme, scoring interactions for optimal orientations, much as it is realized in the LUDI algorithm [22]. Obviously, the algorithm is optimized for high-throughput search, taking into account only the most striking aspects of the pharmacophore. It remains to be seen, whether such minimal representation can account sufficiently for the requirements in affinity and specificity needed for new drugs.

## Bringing Chemistry to Life

Looking at the universities of this world, a decreasing number of chemistry departments can be observed. The primary reason for this is an ever decreasing number of students. Is chemistry going to be obsolete? Surely not! Whatever the research related to molecules is about; chemical synthesis will have to be conducted. The question is, however, where will solutions to problems arising in chemistry come from. Will they be given by chemistry itself, or will physics and/or biology provide them? An answer can be obtained from chemistry's history. In its origin, chemistry was a life science. Being busy with increasingly complicated syntheses, however, many a chemist gradually lost the link to this root. But the situation in cancer, hereditary diseases, and viral infections still requires better means of handling and treating molecules. Especially *in vivo*, molecular interactions need to be controlled more precisely, as can be seen, e.g. in gene therapy.

Where else could be the future direction of chemistry? If you look at the history of carbon-feedstock used by chemical industry, some trend can be observed. In the early days of industrialization in the 19$^{th}$ century, the primary material source was coal. Organic syntheses started with the activation of the C-H bond in acetylene. Later, in the 'golden age of chemistry', which started with Woodward and is to end only now, the primary source was petroleum, and chemical synthesis was mainly about activating allylic and vinylic C-H bonds. A huge amount of phenomena and corresponding explanations have been worked out around these two types of bonds. But the resources of petroleum are limited. Its present double role as carbon feedstock for chemical synthesis and source of energy, petroleum will not be able to maintain for ever. Other materials, preferentially renewable ones, will become more and more important. Already today, methanol is the basic chemical with the largest world production (32 Mio. tons/year). To start chemical syntheses with methanol, biogas, or other kinds of biomass, the alkylic C-H bond is the target of activation. In the past, this type of bond has not been in the focus of interest because it is not so easy to activate as the two other types mentioned. Therefore, future research in chemistry could focus on activation and selectivity in the chemistry of alkylic C-H bonds.

The Bozen 2006 workshop has renewed confidence that chemistry can contribute a lot to all these kinds of problems. In any case, interdisciplinarity will be of ever-growing importance. Let me conclude, therefore, with a citation from the German physicist Georg Christoph Lichtenberg (1742 – 1799) [23]:

"*Wer nur Chemie versteht, versteht auch die nicht recht.*"

## References

[1] Fischer, E. (1894) Bedeutung der stereochemischen Resultate für die Physiologie.-*Ber. dtsch. Chem. Ges*. **27:**3228. Fischer, E. Synthetical chemistry in its relation to biology.*J. Chem. Soc*. **1907:**1749. Koshland, jr., D.E. (1995) The Key-Lock-Theory and the Induced-Fit-Theory. *Angew. Chem. Intl Ed*. **33:**2375.

[2] Moy, V.T. *et al*. (1994) Intermolecular forces and energies between ligands and receptors. *Science* **266:**257 – 259.

[3] Bustanji,Y. *et al*. (2003) Dynamics of the interaction between a fibronectin molecule and a living bacterium under mechanical force. *Proc. Natl Acad. Sci.* **100:**13292 – 13297.

[4] de Gennes, P.-G. (1985) *Scaling Concepts in Polymer Physics.* 2nd Edn, Cornell University Press, Itthaca.

[5] Wallmeier, H. (2000) Model-based data compression, from data compression to information condensation. In: Proceedings of Beilstein Workshop *Chemical Data Analysis in the Large: The Challenge of Automation Age*, May 22nd-26th, 2000, Bozen, Italy.

[6] It should be noted that hydrogen bonds have a covalent contribution. See: Ganty, T.K. *et al*.(2000) Is the hydrogen bond in water dimer and ice covalent? *J. Am. Chem. Soc.***122:**1210 – 1214.

[7] Rao, V.V. *et al*. (1999) Choroid plexus epithelial expression of MDR1 P glycoprotein and multidrug resistance-associated protein contribute to blood-cerebrospinal-fluid drug permeability barrier. *Proc. Natl Acad. Sci.* **96:**3900 – 3905.

[8] Gottesman, M.M. (2002) Mechanisms of cancer drug resistance. *Ann. Rev. Med.* **53:**615 – 627.

[9] Azam, M. *et al*. (2006) Activity of dual SRC-ABL inhibitors highlights the role of BCR/ABL kinase dynamics in drug resistance. *Proc. Natl Acad. Sci.* **103:**9244 – 9249. Krishna, R., Mayer, L.D. (2000) Multidrug resistance (MDR) in cancer: mechanisms reversal using modulators in influencing the pharmacokinetics of anticancer drugs. *Eur. J. Pharm. Sci*. **11:**265 – 283. Hanahan, D., Weinberg, R.A. (2000)

The hallmarks of cancer. *Cell* **100:**57 – 70. Smukste, I., Bhalala, O., Persico, M., Stockwell, B.R. (2006) Using small molecules to overcome drug resistance induced by a viral oncogene. *Cancer Cell* **9:**133 – 146.

[10] For the sake of simplicity, epigenesis is not shown explicitly, since its information is encoded as a modulation of the information encoded in the DNA.

[11] Consciousness is seen as being based on a model on the environment in the brain. A.B. coined the phrase: "Remembering the present". See: Edelman, G.M. (2004) *Wider Than The Sky*. *The Phenomenal Gift of Consciousness*. Yale University Press, New Haven and London.

[12] Ackoff, R.L. (1989) From data to wisdom. *J. Appl. Syst. Anal.* **16:**3-9.

[13] Shannon, C.E. (1948) A mathematical theory of communication. *Bell System Tech. J.* **27:**379 – 423. (1948) *ibidem* 623-656.

[14] Atlan, H. (1987) The self-creation of meaning. *Physica Scripta* **36:**563-576.

[15] Woese, C.R. (2002) On the evolution of cells. *Proc*. *Natl Acad*. *Sci*. **99:**8742 – 8747.

[16] Yáñez-Muñoz, R.J.*et al*. (2006) Effective gene therapy with nonintegrating lentiviral vectors. *Nature Med.* **12:**348 – 353.

[17] Woods, N.B., Bottero, V., Schmidt, M., von Kalle, C., Verma, I.M. (2006) Gene therapy: therapeutic gene causing lymphoma *Nature* **440:**1123.

[18] Mayer, E. (1997) *This is Biology.* Harvard University Press, Cambridge. J.B.S. Haldane, J.B.S. (1947) Boni & Gaer, p. 53. New York.

[19] Hermans, J. (Ed.) (1985) *Molecular Dynamics and Protein Structure.* Polycrystal Books Service, Western Springs. Beverage, D.L., Jorgensen, W.L. (Eds) (1986) Computer simulation of chemical and biomolecular systems. *Ann*. *N.Y. Acad*. *Sci*. 482. Frühbeis, H., Klein, R., Wallmeier, H. (1987) Computer-aided molecular design (CAMD). *Angew*. Chem. *Intl Ed*. **26:**403 – 418. van Gunsteren, W.F., Weiner, P.K. (1989) *Computer Simulations of Biomolecular Systems*. ESCOM, Leiden.

[20] Brandt, A. (1977) Multi-level adaptive solutions to boundary value problems. *Math. Comput.* **31:**333 – 390.

[21] Koehn, F.E., Carter, G.T. (2005) The evolving role of natural products in drug discovery. *Nature Rev*. *Drug Discov.* **4:**206 – 220.

[22] Böhm, H.J. (1992) The computer program LUDI: a new method for the de novo design of enzyme inhibitors. *J. Comput*. *Aided Mol*. *Des*. **6:**61 – 78.

[23] From: The Waste Books of Georg Christoph Lichtenberg.