doi:10.2533/chimia.2011.621

FH HES

Universities of Applied Sciences

Fachhochschulen – Hautes Ecoles Spécialisées

Applied R&D at the Universities of Applied Sciences

Urban Frey*[a], Christian Hinderling[b], Olivier Naef[c], and Patrick Shahgaldian[d]

Correspondence: Prof. U. Frey[a], Tel.: +41 27 606 86 57, Fax: +41 27 606 86 15, E-mail: urban.frey@hevs.ch, [a]HES-SO Valais, University of Applied Sciences Valais, Institute of Life Technologies, Route du Rawyl 47, CH-1950 Sion 2; [b]ZHAW Zürcher Hochschule für Angewandte Wissenschaften, Life Sciences and Facility Management, Institut für Chemie und Biologische Chemie, Postfach, CH-8820 Wädenswil; [c]HES-SO Fribourg, University of Applied Sciences Fribourg, Ecole d'ingénieurs et d'architectes, Institut de Chimie, Boulevard de Péroles 80, CP 32, CH-1705 Fribourg; [d]FHNW, Fachhochschule Nordwestschweiz, Hochschule für Life Sciences, Institut für Chemie und Bioanalytik, Gründenstrasse 40, CH-4132 Muttenz

Abstract: The four institutes of the Universities of Applied Sciences working in the field of Chemistry and Life Sciences present their activities with selected projects.

Keywords: Antibiotic · Applied research · Biosorption · Calixarene · Cell culture · Hematite nanoparticles · Micropollutants · Microreactor · Nanofibers · Nanomaterials · Pheromones · Phosphate recycling · Waveguides · Universities of Applied Sciences

Introduction

The Universities of Applied Sciences are practically-oriented education and research establishments. Two priorities are defined in accordance with the 'Master plan for the Universities of Applied Sciences 2004–2007': the guarantee of a high-quality education and applied research and development. This specific mission for Universities of Applied Sciences has led them to work closely with academic and commercial organizations.

According to the reference document by the Rectors' Conference of the Swiss Universities of Applied Sciences, their applied research and development (aR&D) is used to designated all activities which have as their main aim the creation of new knowledge and the combination of different aspects of existing knowledge in order to valorize it as promptly as possible in academic or practical environments. Hereafter, each institute highlights three examples to show its specific aR&D activities.

EIA-Fribourg

The Institute of Chemistry at the Ecole d'ingénieurs et d'architectes de Fribourg (EIA-FR) has core competences in chemical process research and development up to the production of kg-quantities of complex small molecules. The team develops efficient and robust syntheses of a given molecule based on cleantech solutions within a sustainable development vision. This includes thorough analytical work (*e.g.* off-line and on-line in-process control (PAT); product characterization), physicochemical process characterization regarding thermal safety and kinetics (*e.g.* RC1, DSC, TGA), synthesis work (route finding, route enabling and optimization) and engineering aspects based on new technology platforms, supported by state-of-the-art tools (*e.g.* transfer to continuous processes, distillation & filtration, chemical engineering, and automation). Our competences are in the fields of catalysis, polymers, oils, fats, nanotechnology, fine chemistry, and pharmaceuticals.

Grignard Reaction with Microreactor

During his Bachelor work, Mathieu Roch realized a Grignard reaction under the supervision of Prof. Ennio Vanoli and Prof. Olivier Naef. 3-Butenylbenzene was synthesized from (chloromethyl)benzene and 3-chloro-1-prop-ene. Fig. 1 shows the tube for the preparation of the organo-magnesium, including a temperature sensor for system control. By replacing the PTFE tube with stainless steel the reaction showed different reaction rates in function of time, probably due to the inhomogeneity of the heat transfer involving different temperatures in the tube profile. Using on-line infrared spectroscopy highlighted the different production mode as a function of the material choice. The study of this reaction with this experimental set-up ensures better temperature control and greater selectivity.

Fig. 1. PTFE tube for the preparation of the organo-magnesium (source: TB2010, Criblage de réactions à l'aide d'un microréacteur, Mr. Roch)

Synthesis and Transformation of Hematite Nanoparticules

Changing the properties of a given inorganic–organic hybrid combination in material science is often achieved by modifying the concentration of the integrated particles. This can be disadvantageous leading to dramatic changes in the processing conditions. Among iron oxide particles, hematite particles (α-Fe_2O_3) can be synthesized in a desired anisotropic shape, such as spindles, by wet chemistry methods such as the forced hydrolysis of $Fe(ClO_4)_3$. However, hematite nanoparticles exhibit only weak ferromagnetic properties. Hence, hematite synthesis is followed by a reducing step under hydrogen flow to produce magnetite particles (Fe_3O_4). Magnetite particles offer a stronger magnetic response compared to hematite ones, however the process for the reduction must be carried out under continuous flow of hydrogen at high temperature which poses obvious safety issues. Synthesis of hematite nanoparticles at intermediate scale in a 15 L batch resulted in 15 g of spindle-shaped nanoparticles with a long axis of 284 ± 45 nm and a short axis of 51.8 ± 7.1 nm. Subsequently we developed an innovative autoclave system for the reduction of hematite into magnetite in a closed safe environment. Study of the crystalline structure using XRD pattern analysis shows that reaction at 300 °C/11 bars for 30 hours resulted in almost pure magnetite with almost no change in the spindle-type morphology of the nanoparticles. The first results obtained during this on-going work pave the way for the development of a versatile and controlled transformation process of hematite spindle-type nanoparticles in autoclave conditions, under hydrogen gas pressure, maintaining the anisotropic particle morphology and tuning crystalline structure.

Synthesis of Male Mouse Pheromones

Researchers from the Department of Pharmacology and Toxicology of the University of Lausanne recently approached us regarding the possibility to provide them with mouse pheromones. Dr. Broillet and Dr. Brechbühl needed small amounts of two male mouse pheromones, exo-dehydro-brevicomin and 2-sec-butyl-4,5-dihydrothiazole, in order to validate a new imaging protocol on living tissue slices of the mouse vomeronasal organ, the olfactory subsystem responsible for detecting pheromones.

After a review of the literature regarding the synthesis of the two compounds, we proposed racemic syntheses based on known procedures. Chiral syntheses were also reported for the two pheromones but they are far too complex and the planned physiological experiments could also be done with racemic material. A sample of 130 mg of exo-dehydro-brevicomin was prepared from commercially available exo-brevicomin by bromination followed by an elimination reaction. For the key step of the synthesis of 2-sec-butyl-4,5-dihydrothiazole, the formation of the dehydrothiazole, there was a need for some optimization work, especially on the product isolation and purification. We were finally able to prepare 3.4 g of pure 2-sec-butyl-4,5-dihydrothiazole. An important aspect within this project was good communication and experimental planning between the chemists and the physiologists as the products had to be delivered on time when the mice had reached the age for testing. We were made aware that we had to pay special attention when handling these biologically active compounds, as they are, for example, light sensitive and extremely volatile. These compounds are active at low concentrations (10^{-12} M) inducing specific innate behavior. The two male mouse pheromones were used successfully at the Department of Pharmacology and Toxicology. They induced intracellular calcium changes in mouse vomeronasal neurons indicating neuronal activation and finally allowed the validation of the model system to test and identify new pheromonal ligands.

This project demonstrates the competences in organic synthesis and analytics of complex molecules and also the benefits of local cooperation between 'non-chemical' universities and FHs.[1]

FHNW Muttenz

The competencies and capabilities of the Institute of Chemistry and Bioanalytics (ICB) at the School of Life Sciences of the FHNW cover major fields in Life Sciences. In biochemistry, bioanalytics and diagnostics, our teams work on the development of biosensors, new diagnostic assays and cellular test systems for applications in preclinical drug development and drug metabolism and toxicity. The nanotechnology Group of the ICB is experienced in characterizing nanomaterials by all means of analytical, microscopic and surface analysis technologies. It is also active in the synthesis of nanoparticulate materials (organic, inorganic and hybrid) and in surface modification. Hyphenated-MS-MS technologies and NMR-spectroscopy are core competences in our instrumental analytics group. These techniques are applied for the analysis of pharmaceuticals and for the structural characterization both of small molecules and proteins. In chemistry we have a strong expertise in organic synthesis and supramolecular chemistry. Our synthetic programs range from multistep syntheses of small molecules for Medicinal Chemistry, diagnostic tools compounds based on peptides and carbohydrates to the preparation of new materials used in supramolecular recognition. The chemical engineering group covers process development and optimization in the multi-kilograms scale and downstream processing with a special focus on membrane technology.

Nanomaterials with Selective Molecular Recognition Properties of Active Pharmaceutical Ingredients (APIs)

The preparation of nanomaterials possessing specific molecular recognition properties is essentially driven by the possibility to enhance the number of recognition sites of the material because of the high surface-to-volume ratio that these materials offer. Among the different possible strategies to design such nanomaterials, our focus is mainly on molecular imprinting and on the use of supramolecular systems to design this kind of nanomaterials (Fig. 2). In the framework of a collaborative work between the Institute of Chemistry and Bioanalytics (ICB-group Shahgaldian) and the Institute for Ecopreneurship (IEC-group Corvini), funded by the Swiss Commission for Technology and Innovation CTI, these two approaches have been successfully applied to different APIs such as paracetamol, aspirin and levofloxacin.[2]

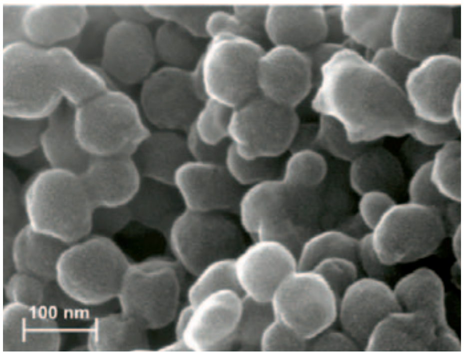

Fig. 2. Scanning electron micrograph of polymeric nanoparticles possessing API binding properties.

Enzyme-modified Nanoparticles for the Elimination of Micropollutants from Waters

The elimination of persistent chemicals during wastewater treatment is a current global challenge. Recent progress in nano(bio)technology offers hope and promise for new developments in catalysis and environmental applications. In a collaboration between ICB and IEC, we have developed an approach based on the use of cheap and resistant oxidative enzymes, *i.e.* laccases, immobilized at the surface of nanoparticles. It has been shown that these systems efficiently catalyze the degradation of bisphenol A.[3,4]

Calixarene-based Solid Lipid Nanoparticles (SLNs) as DNA Cargo Systems for Gene Delivery

The future successes of gene therapy are intimately linked to the development of carrier systems able to transport efficiently a nucleic acid, encoding a therapeutic gene of interest, in the organism to the targeted nuclei, resulting in the expression of the transgene. We have recently developed a novel class of carriers based on cationic calixarenes and demonstrated that these systems can be loaded at their surface, *via* an iterative layer-by-layer process with DNA. These systems have been shown to transfect mammalian cells.[5,6]

HES-SO Sion

The activities of the Institute of Life Technologies are focused on the strategic area of the development of products, analytical technologies and methods in the fields of chemistry, biotechnology and food technology. Proven technical and scientific skills are provided by a team of motivated and multidisciplinary staff; along with high-tech infrastructures we guarantee high-quality services. Therefore, the Institute is in a position to address complex issues in a comprehensive and coherent manner, in a 'value chain' perspective.

Microbial Fuel Cell propelled Phosphate Recycling[7]

Phosphate recycling from digested sewage sludge is an important economic and ecological issue. Today, phosphorus is extracted from daylight mines mainly located in the USA, in China and in Morocco. The demand is ever-increasing whereas the mineral sources are becoming depleted. An important phosphate sink is sewage sludge. However, the distribution of phosphate-containing digested sewage sludge on farm land is considered hazardous and legally forbidden, due to the presence of heavy metals. A series of phosphate recycling methods has been developed in recent years. Most of them use waste water as resource.

The purpose of this project is to extract the phosphate from digested sewage sludge in order to reuse it as fertilizer. The energy for the process is directly extracted from waste waters and recovered using microbial fuel cell technology. At ambient temperature, in the presence of protons and electrons supplied by the metabolic activity of microorganisms, the insoluble $FePO_4$ is reduced to soluble ortho-phosphate, which is then transformed into the fertilizer struvite (Fig. 3). This method eliminates heavy metals and constitutes a promising solution for sustainable phosphate recycling.

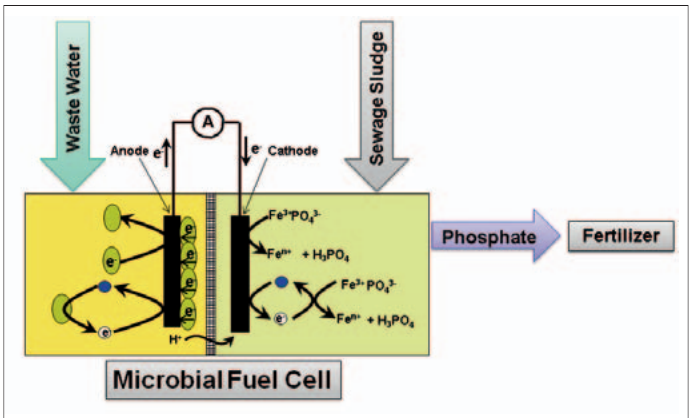

Fig. 3. Microbial fuel cell for the mobilization of orthophosphate from $FePO_4$ contained in digested sewage sludge. Protons and electrons from the cultivation chamber are transferred to the anode. The reduction in the cathode follows two mechanisms: (i) $FePO_4$ containing sludge particles collide with the electrode, (ii) are reached by methylene blue mediators.

STEM-3D[8]

The development of new drugs requires thorough research regarding the pharmacokinetic- and pharmacodynamic toxicity of the active ingredients. In most cases pharmaceutical companies perform animal studies. The costs and the low social acceptance of these studies lead pharmaceutical companies to search for alternatives, such as cell culture or organ models simulating the behavior of complex organisms.

The Institute of Life Technologies participates in a common project of the HES-SO, focusing on the absorption and metabolism of the bioactive substances in the small intestine. For this purpose, colon cancer cells (CaCo2) are placed on permeable membranes. These cells reproduce very fast and are immortal. They behave in a similar way as small intestinal mucosa cells. The substance to be tested is put in contact with the top surface of this cell layer. The substance and their metabolites that pass the membrane are analyzed. This enables an evaluation of the intestinal transfer and the bioavailability of this substance. This model is faster, cheaper and more reproducible compared to animal models.

Biosorption[9]

Micropollutants are substances generated by domestic and industrial activities whose concentrations in the environment, although low, have been constantly increasing over the years. They can be found at every level of the food chain, a problem which regularly leads to a ban on the consumption of certain products.

Biosorption is an adsorption process of solutes from liquid effluents on very low-cost inert biomasses of vegetal or microbial origin. This technique is particularly effective at low concentration, and it is therefore particularly appropriate for processing micropollutants.

The purpose of this project is to evaluate the performances of biosorption in the separation of micropollutants. The point is also to offer techniques for the production and conditioning of biosorbents, notably using encapsulation, so as to enable their use in large scale fixed-bed processes.

Tests have been carried out for treating contaminated effluents, using copper as a model for heavy metals and phenol to represent organic contaminants. The Cu^{2+} ions were adsorbed on powdered seaweed both loose and encapsulated in various hydrogel matrices. Phenol adsorption was carried out with the help of activated carbon of various origins, *i.e.* commercial products as well as carbon produced at the HES-SO Valais and originating from apricot stones.

It was possible to characterize the biosorption process for the two models on various scales, therefore enabling manufacturing, conditioning and implementing of biosorbents.

ZHAW Wädenswil

Coming up with innovative and competitive solutions for problems in applied research and development almost always requires a combination of skills from different fields as well as approaches that go beyond the applications of established technology. This is even more the case for R&D in multidisciplinary fields like the Life Sciences, which are of special significance to the Swiss economy. The Institute of Chemistry and Biological Chemistry (ICBC) of the ZHAW runs an active applied R&D program that is designed to address these needs by integrating competencies in chemistry, biochemistry, chemical- and biochemical engineering, material science, tissue engineering, nanotechnology and state-of-the-art analytical chemistry. In key areas the ICBC is proud of highly specialized know-how, *i.e.* protein purification and protein analysis (C. Zaborosch), tissue engineering (U. Graf), medicinal chemistry (R. Riedl), analysis of volatiles and the chemistry of coffee (C. Yeretzian), technology of filtration (J. Ebert) as well as nanotechnology (C. Adlhart, C. Hinderling).

TEDD, the competence centre 'Tissue Engineering for Drug Development', initiated and headed by Ursula Graf at the ICBC and sponsored by start-up money from the Gebert-Rüf Stiftung aims to bring together interests from academia and industry in these key areas and is an excellent example of this integrative approach.

Key experimental tools at the ICBC include high level bio- and protein analytics using ESI-QTOF-MS and MALDI-TOF MS, PTR-TOF-MS for time-resolved high-sensitivity analysis of volatiles, microscopy techniques including SEM, AFM, confocal Raman and confocal fluorescence, online cytometry and a biochemical engineering lab.

Turning ideas into products is further supported by the on-campus business incubator 'grow' which provides lab space and support for spin-offs and start-ups from within or from outside the ZHAW. The close proximity to the academic environment facilitates mutually beneficial collaborations.

Structure-based Drug Development Applied to the Problem of Antibiotic Resistance

Resistance of bacteria towards known antibiotics is a rapidly growing concern with global impact. The resistance to antibiotics is coded in the genetic material of the microorganisms. Small molecule compounds that suppress the transcription of the segment of the genetic code of microorganisms that code for the resistance (so-called transcription repressor inhibitory compounds, TRICs) are therefore important potential drugs. Within a currently running CTI-funded project, the specialized know how on structure-based drug design and modern synthetic organic chemistry in the group of Rainer Riedl at the ICBC is effectively being combined with the know-how of the ETH-spin off company Bioversys for the rapid screening of potential TRIC compounds. In Rainer Riedl's group, potential lead compounds are identified by docking them *in silico* to 3D-structures of transcription factors (Fig. 4). Identified leads and targeted derivatives are subsequently synthesized at the ICBC and tested at Bioversys. The project has already led to the identification of high-potential drug candidates and a follow-up project is being submitted.

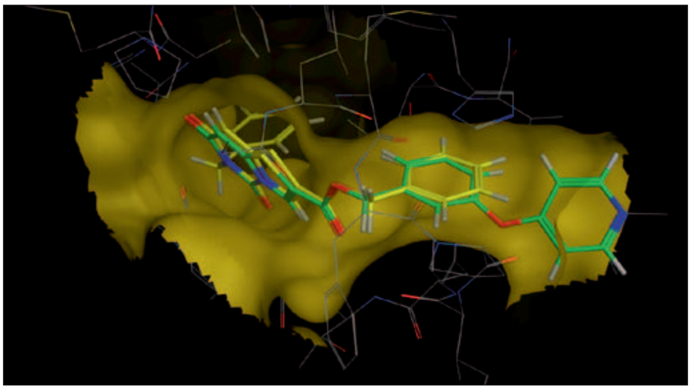

Fig. 4. Structure-based drug design

Creoptix: Grating Coupled Interferometry for Label-free Sensing

High sensitivity label-free sensing as used in affinity studies or in diagnostics is today mostly based on surface plasmon resonance (SPR, *e.g.* Biacore). Interferometric waveguide sensors are known to have the potential to surpass SPR in terms of sensitivity,[10] but have so far suffered from high costs of the complex instruments. Creoptix, a start-up company based in the local business incubator 'grow', has invented a novel sensing scheme that combines the known advantages of low cost planar wave-guides with the high sensitivity of interferometric sensors.[11] The available gain in sensitivity at lower costs enables new applications and markets such as small molecule analysis or point of care diagnostics. In the group of C. Zaborosch, the novel measuring principle is benchmarked with respect to competing principles, and assays and applications for the study of large and small molecules are being developed. The efforts are expected to continue within a CTI funded project.

Functional Nanofibers

Polymer nanofibers with diameters down to few tens of nm and arbitrary length are readily accessible *via* electrospinning and can be obtained as macroscopic nonwoven fiber mats. Such fiber mats combine the huge accessible surface and small diffusion pathways of nanoscale materials with the ease of handling of macroscopic objects and the chemical flexibility of polymers and are therefore of interest in a wide area of applications. At the ICBC we are pursuing applications in biomaterials and as (selective) filter media.

Polymer nanofibers are used as templates in a sol-gel process to obtain nanofibers of titanium dioxide. These are photoactive and semiconducting and show promise in UV assisted sterilization, as self-cleaning filter materials and as electrode materials. In another application we are incorporating molecularly imprinted polymer nanoparticles into nanofibers to obtain capture membranes with high specificity and small diffusion pathways, which show promise for treating fluid streams.[12] Nanofiber-networks are popular as scaffold materials for cells in tissue engineering applications because they form an interconnected network of voids that is very similar in geometry to the extracellular matrix. We are exploring such networks made of protein repellent hydrogels that are selectively patterned with cell recognizable cue molecules as flexible and programmable biomaterials.

Received: June 27, 2011

[1] Roger Marti (Synthesis) and Samuel Roth (Analytical Services) – Direct mandate with Marie-Christine Broillet & Julien Brechbühl, UNIL.
[2] P. Xiao, Y. Dudal, P. F.-X. Corvini, U. Pieles, P. Shahgaldian, *Polym. Chem.* **2011**, *2*, 1264; P. Xiao, Y. Dudal, P. F.-X. Corvini, P. Shahgaldian, *Polym. Chem.* **2011**, *2*, 120.
[3] P. F.-X. Corvini, P. Shahgaldian, *Rev. Environ. Sci. Biotechnol.* **2010**, *9*, 1264.
[4] P. Galliker, G. Hommes, D. Schlosser, P. F.-X. Corvini, P. Shahgaldian, *J. Colloid Interface Sci.* **2010**, *349*, 98.
[5] L. Nault, A. Cumbo, R. F. Pretôt, M. A. Sciotti, P. Shahgaldian, *Chem. Commun.* **2010**, *46*, 5581.
[6] P. Shahgaldian, M. A. Sciotti, U. Pieles, *Langmuir* **2008**, *24*, 8522.
[7] F. Fischer, C. Bastian, M. Happe, E. Mabillard, N. Schmidt, *Bioresource Technology* **2011**, *102*, 5824.
[8] L. Stoppini, W. Andlauer, M. Geiser, H. Keppner, J. Tuberosa, P. Passeraub, H. Eusèbe, Project Call HES-SO N° 23833, **2009**.
[9] S. Crelier, F. Zonnevijlle, Real-Tech HES-SO funding No 20912, **2008**.
[10] K. Schmitt, B. Schirmer, C. Hoffmann, A. Brandenburg, P. Meyrueis, *Biosensors and Bioelectronics* **2006**, *22*, 2591.
[11] P. Kozma, A. Hamori, S. Kurunczi, K. Cottier, R. Horvath, *Sensors& Actuators B*, **2011**, *155*, 446.
[12] R. Büttiker, J. Ebert, C. Hinderling, C. Adlhart, *Chimia* **2011**, *65*, 182.