

MECHANICAL ENGINEERING | PHYSICS |
PRESERVATION OF THE ARCHITECTURAL
HERITAGE | STRUCTURAL, SEISMIC
AND GEOTECHNICAL ENGINEERING |
URBAN PLANNING, DESIGN AND
POLICY | AEROSPACE ENGINEERING
| ARCHITECTURAL COMPOSITION |
ARCHITECTURE, BUILT ENVIRONMENT
AND CONSTRUCTION ENGINEERING |
ARCHITECTURAL, URBAN AND INTERIOR
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ELECTRICAL ENGINEERING | ENERGY AND
NUCLEAR SCIENCE AND TECHNOLOGY |
ENVIRONMENTAL AND INFRASTRUCTURE
ENGINEERING | INDUSTRIAL CHEMISTRY AND
CHEMICAL ENGINEERING | INFORMATION
TECHNOLOGY | MANAGEMENT ENGINEERING
| MATERIALS ENGINEERING | MATHEMATICAL
MODELS AND METHODS IN ENGINEERING



Coordinator:
Prof. Alessio Frassoldati

DOCTORAL PROGRAM IN INDUSTRIAL CHEMISTRY AND CHEMICAL ENGINEERING

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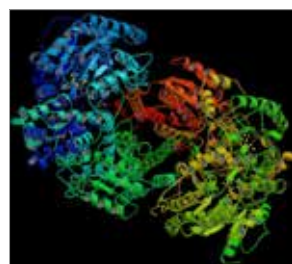
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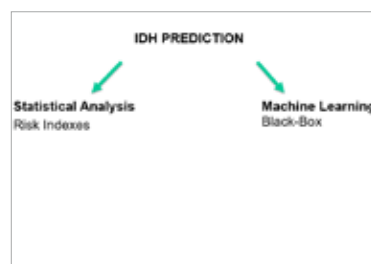
Grants

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The Doctoral Program in Industrial Chemistry and Chemical Engineering (CII) is designed for students aiming to get a deep expertise on chemical processes and on material processing either as the inherent research or in the process design and development activities. The program is the ideal extension of the Laurea Magistrale degrees in Chemical Engineering, Safety and Prevention Engineering, Materials Science, Material Engineering, Industrial Chemistry and Chemistry, but it is also open to graduated in other scientific disciplines.



Structure simulation of
Nitrogenase enzyme for N_2
conversion to NH_3

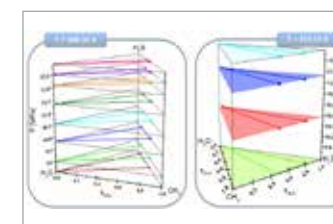


Amyloid peptide nanohelices in
solution as ordered nanomaterials
(Nature Commun. 2015, 6:7574, DOI:
10.1038/ncomms8574)

The general topic for the doctoral program in Industrial Chemistry and Chemical Engineering is the application of the chemical and physical

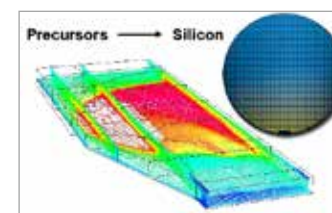
knowledge to all the activities related to the synthesis, design, production and transformation of chemical substances and materials (like advanced inorganic, polymers and biomaterials). These studies involve not only the synthesis processes but also the related plants, here analyzed starting from the laboratory tests and the pilot plant experiments up to the industrial size ones, always including the careful evaluation of the related energetic, safety and environmental issues. The final goal is to provide to the PhD student the tools and the skills: (1) to design and manage industrial processes at any size scales; (2) to develop new technical applications and (3) to create and to characterize new products and services. The **CII** program covers three years for an overall amounts of 180 credits. The responsibility of the organization and of the contents of the doctoral program is attributed to the Professors Committee (PC). At the beginning of the Doctoral program, a tutor and a thesis advisor is assigned to each student. The tutor has a supervisor function during the whole doctoral program, whereas the thesis advisor is responsible for the thesis work.

"Biocompatible polymer nanoparticles
functionalized with a fluorescent dye
for in vivo imaging studies"



VLE and VLLE measurements for the
system $CH_4-H_2S-H_2O$.

The PC defines a specific curriculum for each student, on the basis of the tutor suggestions. The curriculum has to be designed in order to both cover deficiencies in the student scientific formation and to give a high-quality technical and scientific preparation. Since 2001 (XVII cycle) the **CII** program graduated more than 100 students, being ~80% now working in industry and the remaining in the university or government research centers. These data demonstrate the great link existing between the performed researches and the industry. In fact, during the years, numerous the research topics were directly supported by industrial companies, like ENEL, LPE Epitaxial Technology, Bracco, Flamma, Mapei, Solvay Specialty Polymers, RSE, Isagro, Tecnimont, Biochemtex, Pirelli.



Simulation of a chemical vapor
deposition reaction for silicon films
deposition



Hazardous gas dispersion simulation
from an industrial accident

STRATEGIES TO EXPLORE ESTABLISHED POLYMERIZATION TECHNIQUE FOR NOVEL APPLICATIONS

Azzurra Agostini - Advisor: Prof. Davide Moscatelli

Polymeric materials represent a promising strategy for a variety of fields, including the delivery of different kinds of drugs in many therapeutic applications and the formulations of additives for the oil and gas companies. During the last few decades, the attention on new materials has been growing due to the possibility to have biodegradable and/or biocompatible products. The number of laws and regulations has been deeply increased to promote the development of novel environmental friendly materials, also suitable for the human uptake. In this work, the well-known Free Radical Polymerization (FRP) has been exploited for the problem of fluid losses during drilling operations in the extraction of the oil and in the synthesis of polymeric carriers for the delivery of drugs in the gastrointestinal tract.

FRP is a widely used polymerization technique in industry for its many advantages, such as the relative easiness of implementation, the good batch-to-batch reproducibility, the tolerance of radicals towards functional groups and the possibility to use water as solvent. The kinetic scheme involves three steps: the initiation, where radicals are generated, the propagation, in which the polymeric chains

grow, and the termination, that occurs when two radicals combine together. This study focuses more in detail on the initiation, which allows the formation of free radicals starting from an unstable compound through a slow reaction. This usually occurs with the homolytical cleavage of bonds which can be thermal, chemical or by radiation. In this work, only the thermal and the redox generation of radicals have been considered. The best initiator must be chosen according to the operative conditions of the process. The safety concerns justify the use of the redox initiation in the solutions developed for the formulation of new additives for the drilling operations. In this case, the beginning of the reaction is based on a redox system that generates the heat wave that triggers the decomposition of the initiator which starts the polymerization

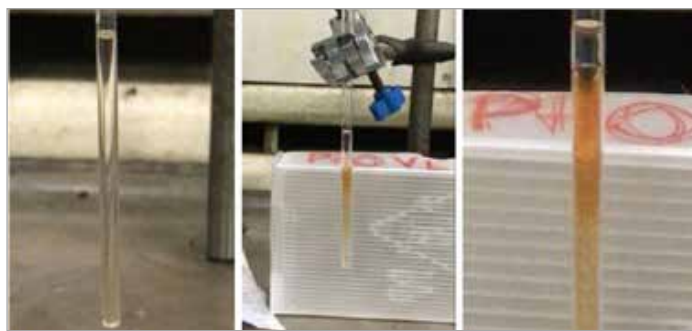


Fig. 1 - Evolution of the frontal polymerization over the time.

(Figure 1).

This kind of FRP is called frontal polymerization. As the initiation step involves one oxidant and one reducing agent, it is necessary to formulate a bicomponent mixture. The idea is to start the reaction only when two mixtures are in contact, the reaction must be able to propagate in the direction of the front. This is the strategy adopted to increase the stability of the solution: the reaction occurs only when the two mixtures are put together, avoiding possible unwanted polymerization in the storage, giving the possibility to initiate the reaction even at lower temperature. The thermal initiated FRP is not suitable for the problem of the fluid losses for the impossibility to have a good control on the reaction. In addition, in order to avoid the dilution of the reactive mixture, the

immiscibility of the additives with the mud has been considered. For this reason, two different recipes have been developed according to the polarity of the muds: a hydrophilic monomer mixture was formulated for the oily mud, while a hydrophobic monomer mixture was prepared for the water based mud. The operative constraints were completely fulfilled after the optimization of the formulations taking into account all the degrees of freedom of the system, such as the types and the amount of monomers, cross linking agents, initiators and solvents. The advantages of this strategy, compared to the existing formulations, like concrete, are the rapid restart of operations, the easy injectability, the better control of the operations, the efficient

formation of solid block, and also its reversibility, due to the possible destruction of the hard material.

On the contrary, FRP has been used to synthesize 406 polymers for the applications in the gastrointestinal tract. Due to the versatility of this technique, a wide range of monomers (Figure 2A) has been adopted to obtain a huge variety of materials with different properties and features (Figure 2B).

The chosen 28 monomers have been used for building a polymer library that could have potential applications for gastrointestinal drug delivery. Frontal polymerization is not feasible for this kind of use because of the high temperatures registered in

the first minutes of the reaction. Monomers containing methacrylate and acrylate moieties were selected due to the commercial availability of a wide range of compounds with these substructures and their capacity to form polymers. A library of 406 homo and copolymer has been synthesized using the FRP to develop novel polymeric formulations for applications along the gastrointestinal tract. Additional functionality, such as triggerability, could be engineered into these monomers through a proper esterification reaction. The possibility to customize the property of the final material considering the structure of the starting monomer has been studied for the application of new enzyme-bacteria sensitive formulation, very important for the inflamed bowel disease and colon alteration, where there are lipase and azo reductase enzymes capable to break down the polymer molecules allowing the release of the drugs, like dexamethasone and mesalamine. High-throughput methods have been developed to characterize some of the properties of the polymers, like solubility and degree of swelling.

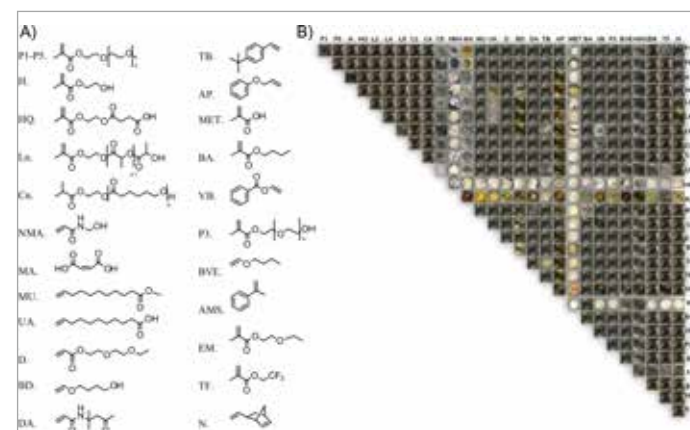


Fig. 2 - Polymer library for gastrointestinal applications.

EVALUATION AND DEVELOPMENT OF BIOMASS CONVERSION PROCESSES FOR THE CHEMICAL AND BIOCHEMICAL VALORIZATION OF LIGNIN

Chiara Francesca Carrozza - Supervisor: Prof. Citterio Attilio

Tutor: Prof. Brenna Maria Elisabetta

Through the integration of green chemistry into biorefineries and the use of low environmental impact technologies, future sustainable production chains of biofuels and high value chemicals from biomass could quickly forecast. Despite the fact that a massive amount of lignin is obtained from the biorefineries, the use of lignin in real industrial processes is far to be obtained. Hence, approaches to develop lignin conversion into high-value co-products are progressively investigated in the attempt to improve the economics of the biorefineries.

In this sense, the PhD project deals with the valorization of biorefinery lignin as by-product from second-generation bioethanol production involving either a biochemical or chemical treatments. The starting materials were obtained from different stages of the cellulosic ethanol plants powered by Proesa™ technology and operated in Crescentino (Italy). More specifically, the research activity was divided in four main Chapters:

Chapter 1: Introduction

In order to face the challenges of becoming independent of crude oil and switching to a more sustainable and carbon neutral

society, biomass has emerged as most prospective raw material in the next future. The basic knowledge about plant origin materials, with specific attention to lignin component will be introduced in this chapter. In this sense, a deep understanding of biomass as renewable feedstock will be provided. Then, the sustainable use of lignin, as the other major constituent in biomass and its potential as starting material for the production of chemicals due to its aromatic structure, will be investigated. Several approaches either focus on the direct utilization of lignin with suitable modification to find application in low value products including dispersant, binders, emulsifiers and resins, or target the depolymerization of lignin macromolecule into high value aromatic fine chemicals will be presented. Finally, a general overview of Crescentino plant and its technology will be reported in order to understand where the raw materials used among all experiments come from.

Chapter 2: Lignin isolation and characterization

Starting from protocol procedures, optimization of extraction and purification treatments combining mechanical treatment (ball milling)

and an intensive washing step will be performed. All the materials were fully characterized by means of different spectroscopic techniques and their properties compared with the raw material. It was found that soxhlet extraction as washing procedure will lead to a material containing 96% of lignin. Meanwhile, in collaboration with the University of Pavia, the effect of the growth of two fungal species, *Trichoderma asperellum* EVT4 and *Pleurotus ostreatus*, which respectively are able to degrade polysaccharides or lignin aiming the biodegradation of the material, was tested. It was found that *Trichoderma asperellum* EVT4 is able to purify the material lowering the content of sugar (reducing sugars or glucose) in a strong way but leaves a complicated tri-dimensional structure. Comparison between chemical and biochemical purification have been done and it can be assumed that both procedure can purify lignin rich residue with a good degree of lignin purity but with different structural properties. Finally, in collaboration with Lawrence Berkeley National Laboratory, a complete biorefinery process (pretreatment, hydrolysis and fermentation) in lab scale using ionic liquid as solvent to degrade and solubilized two

different feedstocks, switchgrass and poplar, was performed. Compared with the traditional pretreatments, IL pretreatment is a relatively new approach and has several parameters that need more investigation. The main drawbacks in ILs pretreatment consist in the extensive washing procedure of the pretreated biomass to remove residual amounts of ILs that may inhibit downstream saccharification and fermentation. To overcome this issue, recently a new wash-free approach was developed. It is based on a one-pot system where IL pretreatment, saccharification and fermentation followed by direct extraction of sugar and recovery of lignin will take place simultaneously. The one-pot process performed gives us good result in term of ethanol yield since the whole hydrolysed glucose was converted. Hence, it was demonstrated that IL pretreatment improves the "delignification" process and significantly increases the enzymatic hydrolysis efficiency. Physical and chemical alterations in lignin structure were investigated with the aim to understand its reactivity during the whole IL treatment.

Chapter 3 and Chapter 4: Catalytic oxidation of lignin and lignin depolymerization by hydrogenolysis.

Since lignin is the richest source of renewable aromatic compounds on earth, its conversion into well-defined aromatic chemicals was deeply investigated by either oxidative and reductive approaches in mild conditions of temperature and pressure. As it is known in literature, the recalcitrance of lignin and its ability to recondensate represent a challenge that inhibit an efficient depolymerization. In the first case, two different catalytic conditions were tested on the purified material by using CuSO_4 or $\text{Co}(\text{salen})$ as catalysts. With copper sulphate the three main aldehydes (vanillin, siringaldehyde and p-hydroxy aldehyde) were produced at pressure equal to 3 bar with an overall conversion of about 5% (coherent with literature results). Instead by using Co complex, p-benzoquinones were formed (mainly methoxy benzoquinone and dimethoxy benzoquinone) with a total yield of 7.2% at 3.5 bar. On the other hand, the purified materials were depolymerized by catalytic hydrogenolysis in alcoholic media with a combination of palladium on

carbon and/or nickel(II) acetate as catalyst. To increase the efficiency of the reaction, *in-situ* preparation of the nickel-based catalyst was carried out by adsorption of metal salt on the material. In 1,2-propanediol as solvent at 210 °C, up to 80% of the material was solubilized with an overall yield of reduced monolignol derivatives of about 18%. Depolymerized lignin fragments (both the liquid residue and the solid residue recovered after precipitation with water) were detected by MALDI-ToF analysis with a molecular weight between 400-1400 Da. To this end, catalytic depolymerization reaction through hydrogenolysis was performed on the one-pot ionic liquid residues. Different conditions of temperature and pressure were tested: at 200 °C the alcoholic solvent (1,2-propanediol) acts as source of hydrogen, while at 90 °C and 140 °C the reaction was performed adding 20 bar of H_2 . Solid residues, from either switchgrass or poplar, reach a maximum value of about 35% but the distribution of product is opposite: about 60% of S unit in switchgrass residue and about 60% of G unit in poplar residue.

EXPLOITING THE GENERALISED SUBSTITUENT EFFECT: THEORY, KINETICS AND APPLICATIONS

Francesco Distante - Supervisor: Prof. Francesco G. Gatti

The research activity reported in my Ph.D thesis provides an overview of the main chemical advantages exploited by the substituent effect in ring-opening and ring-closure processes. In particular, the *gem*-disubstitution effect, the *vic,vic*-trimethyl lock effect and the *vicinal* effect were analysed together with their possible applications. Moreover, a particular focus was fixed on the ring-opening polymerisation (ROP) process, as an alternative polymerisation route, towards the synthesis of more "green" materials.

To start with, the acid catalysed ring closure of substituted γ -hydroxyesters to give lactones was studied. Intramolecular reactions affording cyclic compounds are key-transformations of organic chemistry. To understand better the factors that control the reaction rate of this important class of reactions, a $^1\text{H-NMR}$ kinetic study of a set of monomethyl and/or *gem*-dimethyl substituted esters in CDCl_3 was carried out. We evaluated the effect of leaving group (ethyl vs *i*-propyl ester) and the catalyst efficiency. It was found that: *i*) a monomethyl substitution produces a lowering of the energy barrier similar to that

of a *gem*-dimethyl substitution (Thorpe-Ingold effect); *ii*) the ring closure of *i*-propyl esters is slower than that of ethyl esters; *iii*) strong acids are more efficient than weak acids according to the Brønsted relationship, and *iv*) the Thorpe-Ingold effect is not just an intrinsic feature of the linear precursor but it depends by the catalyst as well. The reaction catalytic cycle was analysed by density functional theory (DFT) computations. The results of this analysis show that the *generalised substituent* effect (mono-, di- and trimethyl substitution together with the leaving group) can be explained in terms of ring strain energy, whereas the Brønsted plot is rationalised in terms of atom charge of electrophile and of distance nucleophile/electrophile.

Another substituent effect, *i.e.* the *vic,vic*-trimethyl lock effect (TML), was investigated to design a paracetamol-based prodrug. A synthetic route was developed to functionalize the drug with a three methyl substituted linear chain. Exploiting the relative stereochemistry of the substituents, it was possible to generate four prodrug diastereoisomers. The latter were kinetically tested in the lactonisation reaction, to experiment a time modulated

drug release in time. Before to proceed with the ring closure process, the prodrug was triggered by the reducing action of the alcohol dehydrogenases enzyme (ADH) and the kinetics studies were carried by means of NMR in D_2O . Taking into account backward studies on a model compound, it was possible to determinate three kinetic rate constants of release (very fast, very slow and intermediate release), to be compared to the observed curve, relative to the diastereoisomer mixture. The experimental curve clearly showed a specific trend, different with respect the calculated curves. Therefore, it was possible to conclude that the drug release may be modulated, undergoing a fast or slow release, depending on the relative stereochemistry of the substituents.

To prove the efficiency of the substituent effect even in ring-opening reactions, a diastereoselective Ring-opening Polymerisation (ROP) of *vic*-disubstituted β -lactone was investigated. A flexible synthesis was designed to functionalize the monomer, generated as a mixture of two diastereoisomers, *i.e.* *cis* and *trans*. Afterward, a screening of different catalysts

was made, both with a cationic and anionic catalytic mechanism. The best catalyst resulted the 1,7,7-triazabicyclo[4.4.0]-dec-5-ene (TBD), which was add into the system together with the initiator benzyl alcohol. A standard polymerisation approach was adapted to react monomer in bulk conditions. Nevertheless, with respect to the general procedures, after polymerisation (24h reaction), the terminal hydroxyl group of polymer was capped using a silylating agent. This step kept the polymer more lipophilic and stable, therefore, the isolation was easier. The resulted molecular weight was analysed by means of NMR, GPC and MALDI-TOF. All the analysis confirmed that *vic*-disubstituted lactones undergo polymerisation with a completely different reactivity and kinetic, depending from the relative stereochemistry of the substituents.

Therefore, it was claimed that the introduction of stereochemical informations in ROP could represent a promising route for the development of new polymers, with the chance to exploit new reactivity and material properties.

Following up on the importance of lower environmental impact polymers, the Ring-opening Polymerisation process was

exploited to produce high molecular weight ($> 30 \text{ kg mol}^{-1}$) polyethylene furanoate (PEF) with more sustainable catalysts, as alternative to the most famous polyethylene terephthalate (PET). The synthesis of the cyclic monomers started first with a prepolymerisation between ethylene glycol (EG) and dimethyl furandicarboxylic acid (meFDCA), to give small oligomers. High cycles purities are essential for successful ROP, which can be obtained by distillation-assisted cyclodepolymerisation (DA-CDP) of short PEF prepolymer, comparing different high dilution systems ($10, 20$ and 30 g L^{-1}) in high boiling solvents such as *o*-dichlorobenzene. In the quest towards a more sustainable future, a selection of more biocompatible initiators based on human body metabolism metals such as Zn and Fe is also an interesting choice due to their benign nature. Acetates and alkoxides of these metals present themselves as viable alternatives to traditionally used Sn/Ti/Sb-initiators. $\text{Fe}(\text{OAc})_2$ and $\text{Zn}(\text{OAc})_2$ successfully yielded complete ($> 95\%$) cyclic PEF oligomer conversions at fibre-grade molecular weights ($M_n = 15 \text{ kg mol}^{-1}$). On the other hand, alkoxides of Fe and Zn showed higher activity than their respective

acetates at equal concentrations, and thus increased PEF-ROP rates. However, the most promising results were obtained by tri-functional alkoxides, such as $\text{Fe}^{3+}(\text{OEt})_3$, which proved most reactive and allowed for non-plasticised ROP, yielding above bottle-grade molecular weights (30 kg mol^{-1}).

EXPERIMENTAL STUDY OF THE SEDIMENTATION OF COMPLEX COLLOIDAL SUSPENSIONS

Enrico Lattuada - Supervisor: Prof. Roberto Piazza

The main subject of the PhD project I carried on is particle sedimentation in natural or artificial gravity (i.e. in a centrifuge), a very common process spontaneously occurring in the natural environment and exploited in many industrial applications. Besides, the investigation of particle sedimentation has also been seminal to the development of statistical physics, thanks to the landmark results of Jean Perrin (Nobel laureate in Physics in 1926) which provided the first indisputable evidence of the physical existence of the atoms by giving experimental support to the Einstein theory of Brownian motion. In fact, moving in the footsteps of Perrin, one can take advantage of colloidal suspensions as model systems, where interparticle interactions can be carefully controlled, to obtain further important and general evidence on sedimentation processes. However, in spite of the extensive investigation performed on colloidal systems, several aspects of sedimentation processes are still poorly understood. In my PhD work, I have specifically addressed several open problems related to the dynamics of sedimentation, or in general of the restructuring processes induced by natural and artificial gravity, in complex

suspensions. By this I mean either suspensions where the interparticle interactions are tuned by exploiting the effect brought in by a suitable macromolecular additive, or dispersions where more than a single colloidal species is present. This approach has allowed me to investigate for the first time several topics related to the effects of gravity on colloidal systems. Crucial to perform some of these investigations has been the development of a new optical correlation method. One of the open questions in sedimentation kinetics is how much interparticle interactions affect the sedimentation velocity of a colloidal fluid. To investigate this topic, we exploited the effect of the addition of a surfactant to a model system of hard spherical colloids. In suspension, surfactants organize into globular micelles, themselves in fact little colloidal objects of nanometric size. These induce strong attractions between the colloidal particles due to the so-called “depletion forces”, which have a range given by the micellar size and a strength that increases with increasing surfactant concentration. Usually, a suspension at moderate particle concentration settles slower than an isolated particle, due to the hydrodynamic interactions between the particles (namely, to

the solvent backflow induced by a settling particle, which perturbs the motion of the others). This effect is known as “hindered settling”. We found however that, when the attractive forces induced by depletion effects are sufficiently strong, the settling speed increases consistently, to the point that the settling speed of a moderately concentrated suspension becomes larger than the single particle value, a very peculiar effect that we dubbed “promoted settling”. At high particle concentration, however, the effect of hydrodynamic interactions becomes dominant and the system reverts to hindered settling. This result, obtained for the first time for a simple and well characterized model system, was shown to be relevant also for the investigation of protein association processes. When the strength of the depletion forces is further increased, the colloidal particles undergo a phase separation process with the solvent. Due to the short-range nature of the depletion forces, however, the system does not demix into two phases, but rather gets arrested into a disorder solid phase. This “physical” gel is made of particle clusters linked by relatively weak intercluster bonds, which can easily break and re-form due to thermal agitation. The value

of the particle concentration in the clusters that initially form, and what is the mechanical response of these depletion gels under a compressive stress, is however still debated. We investigated the properties of depletion gels by forced sedimentation, showing that a centrifuge can be actually turned into an instrument capable of performing accurate compressive rheology tests. When more than one colloidal species is present (which is the usual case occurring for instance in the formation of geological sedimentary deposits), the settling kinetics becomes very complicated, so that theoretical results have been developed only for suspensions very dilute in all components. Yet, even in this limiting case, our group discovered an unexpected hydrodynamic instability that leads to what one could call a “settling disaster”: namely, the fastest settling species, instead of settling smoothly, soon develops complex sedimentation patterns that have no theoretical explanation. In my work, I have investigated the settling processes before this instability sets in, showing that the sedimentation velocity of the fastest settling species does not conform at all with the theoretical prediction. Rather, it requires to take into account subtle modifications of the expression for the buoyancy force felt by a particle settling in a “sea” of different particles, the so-called “Generalized Archimedes’ Principle” formerly proposed by our group. A very puzzling question concerns velocity fluctuations in sedimentation. In fact,

hydrodynamic interactions between the particles, because of their long range, lead to a paradoxical theoretical result (“Smoluchowski paradox”): in an unbound fluid the overall effect of the hydrodynamic interaction perturbations leads to diverging settling speed of the suspension. As shown by Batchelor in 1972, this unphysical result can be corrected by carefully taking into account the solvent backflow, which is always present if the container has a bottom. However, the ingenious observation made by Batchelor – which also led him to quantify the effect of hindered settling for hard spherical particles – works only for the average settling speed, and not for the fluctuations of the settling velocity about the mean, which are still predicted to diverge with the container size. The experimental evidence collected so far seems to contradict this result: fluctuations are large but seem to be finite in range and amplitude. However, all existing measurements of the fluctuations of the settling velocity have been performed on suspensions of “non-Brownian” particles, namely large particles that can be directly tracked by Particle Image Velocimetry (PIV) and whose Brownian motion is almost negligible. Yet, this choice generates several experimental problems related to the homogeneity of the initial condition. The aim of my work has been performing a first investigation of sedimentation velocity fluctuations for small, Brownian particles. The results I obtained, although preliminary, show that velocity fluctuations

in suspensions of Brownian particles are drastically different from those observed in the non-Brownian case. Besides, I managed to unravel subtle convective patterns in a settling suspension, presumably due to tiny temperature gradients (of the order of mK/cm), which are arguably the slowest convective effects so far observed and that hint at the existence of a non-equilibrium interfacial tension between a settling suspension and its supernatant (the pure solvent left above by the settling particles). To obtain the aforementioned results required to devise a technique to detect the full velocity field in a suspension of particles that are too small to be optically resolved. To this aim, inspired by some recently developed optical correlation methods, I designed and tested an optical system which extracts the local velocity field from the motion of the “speckle pattern” generated on an image by the interference between the transmitted light and the light scattered by the particles. This setup was also employed to get preliminary insights into the complex hydrodynamic instability in the settling of binary colloid mixtures discussed above.

BIOBASED CHEMISTRY AND TECHNOLOGY: DERIVATIVES OF ALDARIC ACIDS AS KEY INTERMEDIATES FOR SUSTAINABLE PRODUCTS AND MATERIALS

Jiemeng Li - Supervisor: Prof. Attilio Citterio

Considering the fact that humanity has put a significant global impact on our planet, concerning both the geology and environment, national governments and industries have reached a consensus of developing a sustainable economy a resource-efficient, green, and competitive low-carbon. Among all the strategies applied, green chemistry should be and must be a driving force for mitigating pollution and reducing resource consumption, and then reach the longer-term decarbonisation target to fulfil the final goals of the Paris Agreement. In order to achieve this goal, the modern biorefinery need to deeply investigate first the chemistry, involved in the synthesis of platform molecules and then the alternative strategies to convert them into value-added sustainable products and materials by a combination of biotechnological and chemical methods. Different C5 and C6 sugar building blocks and structural polysaccharides containing repeating units of D-glucose, D-galactose, D-mannose, D-xylose, D-arabinose, etc., constitute the major structure of biomass. Among these, D-fructose and D-glucose are economically suitable for use as chemical raw materials, in particular for the production of dicarboxylic acids.

Aldaric acids are a group of sugar acids, where the terminal hydroxyl and carbonyl groups of the sugars have been replaced by terminal carboxylic acids, and are characterised by the formula $\text{HOOC}-(\text{CHOH})_n-\text{COOH}$. They are key intermediates to useful chemical products and biodegradable polymers. D-glucaric acid, one of the more representatives aldaric acids has been deeply investigated regarding its preparation and reactivity, was identified as one of the top 12 platform molecules by the US Department of Energy (DoE) in 2004. Owing the similar structure, other members of aldaric acid family should still have a huge potential to be exploited. This thesis has addressed representative information from literature on the main aldaric acids (glucaric acid, mannaric acid, and galactaric acid) as an introduction on their synthesis, reactivities, application and its potential as platform molecule. Among these acids, galactaric acid (also known as mucic acid) is of particular interest, as it has been reported as potential intermediate for the production of other C6 dicarboxylic acids of industrial importance such as adipic acid, 2,5-furandicarboxylic acid and terephthalic acid. Moreover, despite the multi-chiral

center nature of aldaric acids, its achiral meso form with no optical activity would be beneficial for its preparation and further synthesis as well as working-up procedure. At moment the possible industrial scale production of biobased galactaric acid has been reported in both chemical and biological methods, which could further contribute to decrease the price of this compound to make it economically feasible as a platform chemicals. Our focus on galactaric acid was on attempt to improve the knowledge on the chemistry of aldaric acids by using a compound supplied directly in acid form, through well identified hydrolytic-oxidative bio processes from natural pectin, which are themselves amenable to further upgrading from wasted biomasses. The aim is to exploit the reactivity of this biobased platform molecule in order to develop cost compatible processes for commodity/fine chemicals and novel functional materials, i.e. polymers based on the pyrrole, amide, and ester functional groups. All the facts, along with the limited knowledge of the chemistry and biochemistry of these compounds, have oriented the choice of aldaric acids as research subject for this thesis. The study was centered mainly on galactaric acid as the

more representative aldaric acid. This thesis mainly deals with the selected reactivities of galactaric acid as follows, dehydration of aldaric acids, identifying lactones and unsaturated intermediates involved; reduction of aldaric acid and their unsaturated intermediates; acylation of hydroxy groups of aldaric acids and role of esters in the elimination of acyl groups; role of aldarate salts (both inorganic and organic) in the above mentioned processes; possible efficient transformations into value-added products and materials, including five membered aromatic derivatives of furan and pyrrole, six membered derivatives of pyrenes, amide salts, diamides and polyamides. The new insights gained in this work inspired the development of novel biorefinery pathways and processes to convert galactaric acid into new platform chemicals/intermediates, enabling sustainable biofuel

production from carbohydrate biomass. The study of the possibility of dehydrating galactaric acid to give its corresponding unsaturated derivatives was carried out according to a two-step acylation and dehydroelimination approach in presence of acid/base catalyst, providing access to pyrone derivatives. The 2,5-dihydroxyadipic acid and small amount of mono-reduced product were further synthesized by the hydrogenation reaction of the pyronecarboxylic acid. Protected and unprotected 1,4-dicarbonyl compounds are proved to be relevant intermediates in the dehydration of galactaric acids. They are efficiently converted under mild conditions by amines to pyrrolecarboxylic acid derivatives and unsubstituted pyrroles by decarboxylation under moderate temperatures. A better understanding of the mechanistic details of the formation of pyrroles

from mucic acid and amines is provided. Galactaro-1,4-lactone is prepared by a simple thermal method with dimethyl sulfoxide as solvent in quantitative yields, opening the possibility to become a potential platform molecule. Inorganic and organic salts of galactaric acid 1,4-lactone are easily prepared and isolated under mild conditions. Mono salt mono amides of mucic acid and diamides were synthesized in high yields by treatment of galactaric acid 1,4-lactone with primary amines, which is a good start point for synthesis of homo- and co-polymers by further thermal polycondensation. Galactaro-1,4-lactone is selectively mono-, di- and tri-formylated. This last compound and the corresponding acetylated derivative undergo selective de-acylation by a base to mono-unsaturated lactone. Similar process is observed from other aldaric acid mono and dilactones. These mono-unsaturated lactones are key intermediates for further elimination to di-unsaturated derivatives furan-2,5-dicarboxylic acid under acid catalysis.

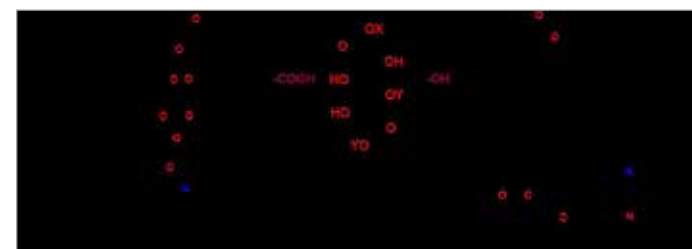


Fig. 1

CVD GRAPHENE FOR ELECTROCHEMICAL BIOSENSING

Lorenzo Pedrazzetti - Supervisor: Prof. Luca Magagnin

Graphene is the first single-atom-thick material ever synthesized. Graphene crystals are monolayers of sp^2 carbon atoms arranged in a honeycomb lattice. Ultra-high tensile strength, exceptional charge carriers mobility, and ambipolarity are only a few of the many properties that earned graphene the attribute of "wonder material". Ever since its synthesis in 2004 a lot of efforts have been made by research group all over the globe to implement graphene properties in consumer technology. Graphene applications span from high strength composite materials to nanosized building blocks for quantum computing, making it not only one of the thinnest materials in nature but also one of the more versatile. The focus of this work is to explore high throughput routes for graphene biosensors production, as well as to develop graphene-based biosensors prototypes. Electrochemical biosensors are becoming more and more important in everyday life, as transducing stimuli from the environment, or the body, and monitoring how these signals evolve in times is a reliable way to improve safety and longevity. Sensors are means of surveying macro- and micro-systems, transforming molecular signals into electromagnetic waves. Graphene offers a valuable

platform to realize cited devices, given its very high surface-to-volume ratio, conductivity, and almost perfect biocompatibility. Graphene can be obtained by top-down or bottom-up approaches: the former involves exfoliating graphite chunks until a single layer is left, while the second requires a metal catalyst, a carbon precursor and the proper thermodynamic conditions. This last process is known as chemical vapor deposition (CVD) and is quite appealing for graphene production at the industrial level. Copper, Nickel, and Cobalt are widely used catalysts for graphene CVD and are also employed as active media in glucose biosensing. Glucose levels monitoring is a very delicate topic as in the next decade glucose metabolic diseases (Diabetes) will be a major cause of death worldwide. The production of Cu, Ni, and Co by different electrochemical processes is hereby studied, in order to design functional catalysts for graphene growth and their use as biosensing electrodes. Copper surface is optimized by applying Hydrogen annealing and electrochemical polishing: in this way, the growth of single-layer graphene by atmospheric pressure CVD (AP-CVD) is achieved. Moreover, Ni is deposited on Silicon by galvanic displacement and used

to grow graphene directly on a semiconductor surface. Then, a novel route to Co electroforming is taken, as a non-aqueous solvent is employed to grow micro-textured Co surfaces. The first characterization of graphene layers on so-produced Co surfaces is presented. Copper substrate appears to be yielding the best graphene quality, hence is used to characterize the hybrid electrode comprised of a composite copper/graphene structure in terms of glucose oxidation electrocatalysis. Pristine graphene is known to be a surface with slow heterogeneous electron transfer (HET) due to its low density of states (DoS) at the interface with the given electrolyte. This is why a surface modification chemical step is implemented to graft graphene surface with oxygen-containing functional groups. The resulting electrode shows improved glucose electro-oxidation performances and good sensitivity (Fig.2). This is due to a bivalent effect of the surface grafting process: on one side, oxygen-containing groups help to increase carbohydrates residence time in the high polarization region right above the electrode surface (double layer), on the other, chemical functionalization of the graphene surface introduces small defects that help to increase its DoS at

the interface. Characterization of such surfaces is done by combining Raman and infrared (IR) spectroscopy. These techniques are complementary, hence, when they are combined, a complete understanding of the analyzed material is obtained. Raman shift is a collection of the light scattered by a surface, while IR spectroscopy yield information about light absorption of a specimen. Raman is of paramount importance in graphene technology, as it is an easy and quick tool to identify single-layer graphene and to diversify films with a different number of layers. Graphene-based DC devices, though, suffer from screening due to the charges accumulated in the double-layer. A brand new prototype for biosensing through microwaves analysis of graphene networks is developed to overcome the 0-frequency limit. To do so, a graphene/gold transmission line is realized via lithography and integrated with a microfluidic structure. The latter is introduced to work as a delivery system for biological media to reach the graphene surface during the network analysis. The proposed platform is employed to monitor the binding of small quantities of single-strand oligonucleotides (Fig.3). Presented work is seminal but make an interesting case of study for graphene-based biosensing. Due to graphene intrinsic small dimensions combined with its useful properties, the development of related nanotechnologies paves the way to the next generation of integrated devices.

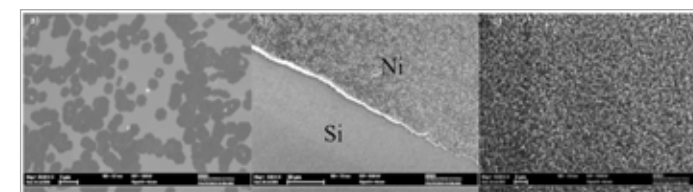


Fig. 1 - SEM images of the various different catalysts after graphene growth, a) Copper, b) Nickel (on silicon), and c) Cobalt

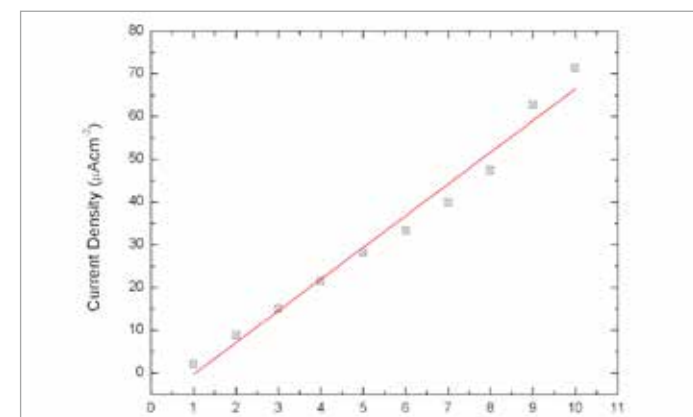


Fig. 2 - Oxygen grafted graphene electrode calibration curve for glucose in alkaline medium

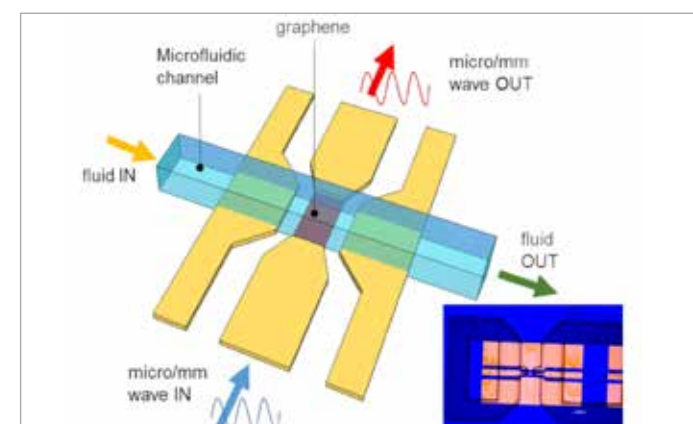


Fig. 3 - Schematic and optical image (onset) of a graphene-based microwave biosensor

SYNTHESIS, CHARACTERIZATION AND BIOLOGICAL EVALUATION OF AMINOGLYCOSIDE-CONJUGATES FOR CELL TRANSFECTION

Chiara Pennetta - Advisor: Prof. Alessandro Volonterio

Gene delivery aims to introduce genetic materials into cells by using vectors for therapeutic purposes. The development of efficient and safe methods for delivery of exogenous nucleic acids into cells, is still a challenging task. Gene transfer by means of non-viral vectors has attracted great interest because non-viral vectors are easy to synthesize, stable, and relatively safe. In this context, cationic lipids and polymers are promising candidates for designing alternative vectors. Cationic lipids and polymers are able to spontaneously complex the genetic material and promote its uptake into cells. However, their clinical application is still hampered by their low transfection efficiency.

Aminoglycosides, which are a class of polyaminosugars with inherent antimicrobial properties, are known to bind to nucleic acids, thus providing a favorable scaffold for the synthesis of non-viral vectors.

During my PhD project I have explored the synthesis and the transfection potential of novel classes of non-viral gene delivery vectors based on the use of aminoglycosides as polar head. Aminoglycosides were tethered to different structures to obtain

non-viral vectors:

- A library of cationic lipids was obtained by exploring a triazine core with different lipophilic moieties in combination with neomycin and paromomycin (chapter II),
- Calix[4]arene, a macrocyclic scaffold already employed in the synthesis of vectors with high transfection efficiency, was also explored in combination with neamine, neomycin and paromomycin (chapter III),
- PAMAM, dendrimeric, well-defined structure that allows for an easy functionalization, was conjugated with neomycin (chapter IV).

All the conjugates were synthesized and characterized through different techniques. Their biological properties as transfectants were also investigated.

The growth of antimicrobial resistance represents a worldwide emergency that need to be urgently addressed. A promising strategy to overcome this threat to human health consists in the improvement of the effect of already existing antibiotics.

During my research project,

I have also tried to enhance aminoglycoside antibacterial activity by synthesizing a library of aminoglycoside-antimicrobial peptide conjugates.

MOLECULAR BASES OF PHOSPHODIESTERASE-4D INHIBITION BY MEMORY-ENHANCING GEBR-LIBRARY COMPOUNDS

Tommaso Prosdocimi - Supervisor: Dr. Emilio Parisini

The neurological mechanisms governing the learning and memory processes depend critically on the cerebral levels of the second messenger cyclic adenosine monophosphate (cAMP) and on the correct functioning of the cAMP/PKA/CREB pathway. In the brain, cAMP levels are regulated by the activity of the type 4 phosphodiesterase (PDE4), an enzyme that hydrolyzes cAMP to 5'-AMP. Consistently, PDE4 is considered an important pharmaceutical target due to its crucial involvement in the signaling of the central nervous system. Indeed, some PDE4 inhibitors (PDE4Is) developed over time have been shown to improve memory and cognitive functions both under physiological and pathological conditions.

PDE4Is have mostly been designed to interact with the catalytic pocket of the enzyme and compete with the cAMP hydrolysis process. As a result, albeit providing interesting pro-cognitive and anti-depressant properties, current standard inhibitors such as Rolipram show also severe side effects due to their lack of isoform-specific binding properties. As many as 20 PDE4 isoforms that are generated by alternative mRNA splicing have so far been identified, each of them having a specific pattern of expression. The long isoforms of

the genes include two regulatory domains, UCR1 and UCR2, while shorter isoforms are characterized by the absence of UCR1 or by the absence of both UCR1 and a portion of UCR2. Moreover, further variability is introduced by isoform-specific N-terminal regions, which are responsible for the interactions with signaling partners. The most extensive structural studies demonstrated that UCR1 and UCR2 mediate enzyme dimerization and that both UCR2 and the CR3 (a third regulatory region) domains are involved in the capping of the catalytic pocket of the enzyme, thus partially blocking access of either cAMP or PDE4 inhibitors. As a result, long dimeric isoforms, and short monomeric ones feature substantially different enzymatic and pharmacologic properties. Burgin et al. demonstrated that it is possible to design allosteric modulators that are partially specific for PDE4D and that are capable of stabilizing the closed conformation of the UCR2 domain of PDE4D over the catalytic pocket through the clamping of a central phenylalanine. Likewise, a similar concept has been suggested also for the design of PDE4B inhibitors that stabilize the capping of the catalytic domain by CR3. Currently, the most promising strategy to obtain a suitable

therapeutic window while avoiding side effects seems to be the design of PDE4D selective inhibitors. Indeed, PDE4D has been demonstrated to be particularly important in memory development and its association with cognition processes has been demonstrated in knock-out mice, which have shown memory improvements similar to those shown by mice treated with Rolipram. Moreover, activating missense mutations affecting PDE4D activity were recently discovered to be associated with acrodysostosis, a rare genetic disease that brings also a phenotype of mental retardation. In an effort to develop a drug that selectively targets the PDE4D isoforms without side effects, a number of compounds, commonly referred to as the GEBR library, that show partial selectivity for the PDE4D isoform have been synthesized. The reference inhibitors of the GEBR library are GEBR-7b and GEBR-32a, which have been shown to improve spatial and objects recognition memory and to increase hippocampal levels of cAMP in transgenic mice. By addressing the biochemical behavior of these compounds both at a structural and functional level, we aim at identifying key structural features that may provide the molecules

with the ability to bind stably into the catalytic pocket while selectively interacting with the regulatory domains of the enzyme. The relatively high number of compounds synthesized to date (up to 90), some of which have interesting pharmacological and toxicological profiles, have so far provided useful, albeit not definitive, information about the inhibitors-enzyme interaction. Therefore, we set out to investigate the structure-function relationship for some selected GEBR compounds and contribute

to the elucidation of the molecular bases of PDE4 inhibition. Our characterization identified interesting structural properties, as well as three major families of compounds that differently dock inside PDE4D catalytic pocket with different conformations: extended, twisted and protruding. Among them, the protruding compounds feature a tail that develops within the S-pocket, pointing towards the external part of the pocket, where a putative interaction with the regulatory UCR2 helix could in principle be exploited. Every

compound has been functionally tested *in-vitro* both on the PDE4D3 long isoform as well as on the PDE4D catalytic domain only, in order to assess whether a differential potency towards the two systems is present. Moreover, the structure of the PDE4D catalytic domain in complex with GEBR-7b and GEBR-32a were used in combination with the most extended X-ray crystal structure of a PDE4B long isoform, in order to investigate the properties of our compounds in a fully-regulated system by means of molecular dynamics simulations. The combination of the structural, functional and *in-silico* information derived from the study allowed us to design the new generation of GEBR molecules there are currently being synthesized in the Bruno's laboratory at the University of Genova.

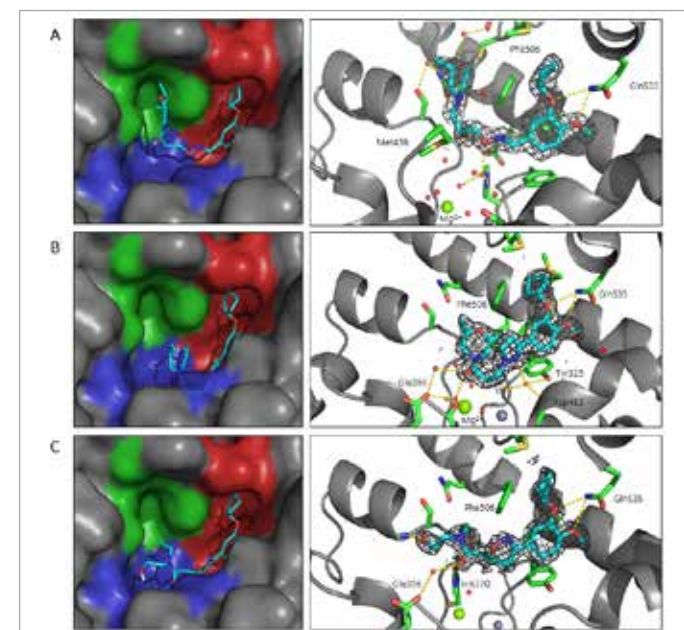


Fig. 1 - Binding conformations of three prototypical members of the GEBR library: (A) GEBR-4a, protruding, (B) GEBR-18b, twisted, and (C) GEBR-26g, extended

ANALYSIS AND DESIGN OF UNCONVENTIONAL EQUIPMENT USING CFD

Matteo Rizzotto - Supervisor: prof. Valentina Busini

In the field of chemical engineering, the knowledge of fluid dynamics is of fundamental importance, since most of the chemical-processing operations present in a plant are carried out in whole or in part in the fluid phase. Examples of this type of operations can be found in biochemistry, energy, materials, oil&gas, pharmaceuticals, polymers, etc. All this emphasis on fluids makes sense especially for two reasons: first of all, under typical process conditions, a huge number of materials exist in liquid or gaseous phase (or they can still be transformed into such phases). Secondly, it is generally more convenient to work with fluids than with solids. With reference to this, even some operations with the solids can be conducted in a “quasi-fluid” manner, such as fluidized-bed catalytic reactors, and the transport of carbon particles in pipes where water is used as a medium for transport and agitation. In order to describe correctly and completely what happens inside an equipment, unfortunately the solution of the fluid flow is not sufficient. The reason for this is linked to the fact that in the process equipment of a chemical plant (heat exchangers, reactors, separation units, pumps, pipes, etc...), the flow is coupled to the transport of mass and/

or energy. While in previous decades empirical correlations and experiments were the most used tools, nowadays these simplifications can be revised or replaced, also trying to understand in detail the coupling between flow, mass, and energy transfer. Industry must invest in the development of new technologies as they allow to reduce costs and increase profits, while ensuring compliance with increasingly stringent regulations. For decades, the aerospace, automotive and defense industries are using simulations for the design and development of new products, as they minimize the costs of physical testing, assessing new concepts and product performance in a low-risk virtual environment. The computational techniques coupled with the most advanced experimental techniques are the most comprehensive and complete tool available to the engineers to assist them during the design and management phases. One of the most used computational techniques is computational fluid dynamics (CFD). Regarding chemical engineering, the use of CFD in the chemical, oil and gas, pharmaceutical, and process industries has been growing as we can see in Figure 1, which is based on research done on the Web of

Science core collection at October 2018. It shows the number of papers related to chemical engineering and CFD over periods of 5 years.

This is partly due to the development of the CFD codes, which are nowadays capable of integrating complex models (multiphase, reactive flows, advanced turbulence models, etc.), and in part to the increasingly available computational resources that allow to obtain results in shorter times. Suffice it to say that the same work that ten years ago required a week, today can be obtained in a few hours of simulation.

In comparison to traditional approaches, which are based on experiments and correlations to take into account non-idealities during project phases, CFD is able to guarantee a more detailed point of view: the calculated solutions provide complete data on fields of velocity, temperature, species, and so on, that engineers can use to better understand the physics of the problem.

This PhD project is focused on the analysis and the design of unconventional equipment using CFD. The CFD code chosen is the commercial software ANSYS Fluent, one of the most widespread programs available in the market. This tool allows to

simulate a wide range of thermo-fluid-dynamics phenomena, providing a comprehensive set of advanced physical models capable of accurately describe fluxes of both compressible and incompressible fluids, heat exchange processes, turbulent flows, multi-phase fluxes and more.

The first part of the project consists of the simulation of non-reactive systems. Two systems will be studied. In the basic case, a static hood developed by Polimi LabOlf was analyzed, in order to understand if the sampling point is representative of the internal conditions of the hood. The following step was to apply the model to a wind tunnel, which was characterized by poor internal fluid dynamics performances, which did not allow to obtain a correct velocity profile on the surface sample, and presented the problem of having to be easily transportable and dismountable for use in the field. The CFD was used to improve its design. In the second case, a full multiphase simulation is performed, with the use of a population balance model to simulate the nanoparticles precipitation in a T-junction microreactor.

The second part of the project consists of the simulation of reactive systems. In the first

section the objective was the CFD simulation of two different reaction accidents with two different failure scenarios (block of the impeller, and loading error), in order to determine the best location of the temperature sensors inside the reactor. In the second section, a new hybrid model capable of handling any type of homogeneous liquid reaction in any condition was developed and validated.

BIODEGRADABLE THERMO-RESPONSIVE POLYMERS FOR DRUG DELIVERY AND TISSUE ENGINEERING

Mattia Sponchioni - Supervisor: Prof. Davide Moscatelli

Thermo-responsive polymers belong to the so-called stimuli-responsive materials and are able to respond to temperature modifications in the surrounding with a sharp and well-defined phase transition. The thermo-responsive polymers characterized by a lower critical solution temperature (LCST) are the most common, especially in aqueous environments, and are soluble in the solvent below the critical temperature while they become insoluble above it. The phase separation occurring at the transition temperature (cloud point) leads to the formation of a polymer-rich phase in water, as the result of the inter- and intra-chain polymer interactions becoming thermodynamically favourable, as clearly visible in **Figure 1**.

Due to their smart behaviour, thermo-responsive polymers are finding growing attention in several fields, such as oil and gas, electronics and chromatography. In the biomedical field, they are regarded as promising materials to achieve complex tasks, simulating the dynamic behaviour of the most important biomolecules that regulate the basic processes of life in response to external stimuli. In particular, the specific behaviour of thermo-responsive polymers is regulated

by their peculiar architecture and composition. Therefore, the advent of controlled radical polymerization techniques (CRP), enabling a precise control over the polymer structure, has greatly contributed to the widespread adoption of thermo-responsive synthetic polymers. In particular, in the biomedical field, these smart materials have paved the access to novel treatments, such as a more selective drug delivery, cartilage regeneration and delivery of genetic material. Of course, in this case, fundamental prerequisites are the biocompatibility and biodegradability of the formulation. Therefore, in this work, the reversible addition-fragmentation chain transfer (RAFT) polymerization and the ring opening polymerization (ROP) were combined to synthesize highly controllable, biocompatible and biodegradable thermo-responsive polymers with a LCST behaviour in water. The possibility of exploiting their peculiar thermal response to achieve complex tasks was then demonstrated by exploring specific applications in the drug delivery and tissue engineering fields, as shown in **Figure 2**.

Focusing on the use of thermo-responsive materials for drug delivery applications, the development of biodegradable

and thermo-responsive nanoparticles (NPs) able to entrap and control the release of a hydrophobic drug is demonstrated. The great advantage brought about by the thermo-responsive behaviour compared to the traditional polymer NPs is to allow for a precise control over the drug release in response to a temperature increase. The drug can be loaded in the NP core, administered to the patient and the drug release induced by applying a mild hyperthermia, achievable via thermal heating or photoillumination, in the target site only. This possibility, combined with an efficient drug retention below the LCST, prevents the often severe side effects associated to a non specific drug release in



Fig. 1 - Visualization of the phase transition for polymers with a LCST or a UCST behaviour.

healthy tissues.

In addition to smart NPs, thermo-responsive polymers can also be exploited to produce injectable hydrogels. These are formulations in a free-flowing state at room temperature and able to form self-standing gels once heated above the LCST. The importance of such formulations relies on the fact that no invasive and expensive surgical operations are required to implant a pre-formed hydrogel scaffold in the patient. On the other hand, with a thermo-responsive formulation, the active principle can be uniformly distributed by simply mixing with the liquid, the solution injected through a needle and then the formation of a gel scaffold induced at the body temperature. This also allows obtaining materials able to adapt to any shape and cavity of the site of action. In this work, thermo-responsive formulations with a reversible sol-gel transition were developed and their efficacy in the delivery of active principles evaluated. In particular, to make this formulation biodegradable, the synthesis of a macromonomer

comprising an oligo(ester) chain functionalized with a methacrylate group was synthesized via the ROP of ϵ -caprolactone using 2-hydroxyethyl methacrylate (HEMA) as initiator. This macromonomer was copolymerized with thermo-responsive monomers already known in the literature to obtain smart systems that can be hydrolysed under physiological conditions. These copolymers can form a gel above the LCST of 30 °C. However, the hydrolysis of the lateral oligo(ester) chains incorporated in the copolymer increases their overall hydrophilicity during time, and in turn the LCST. Once this phase transition temperature overcomes the body temperature, the reverse gel-sol transition is experienced and the copolymers return completely soluble and can be easily eliminated through the kidneys.

Finally, a method for the facile and cost-effective deposition of thermo-responsive polymer coatings on tissue culture polystyrene (TCPS) surfaces is

developed. This finds application in the smart cell harvesting. Cells in fact were demonstrated to adhere to hydrophobic surfaces, while spontaneously detach from hydrated substrates. Here we show how the thermo-responsive transition can replace the traditional enzymatic treatment to achieve an efficient and non-detrimental cell harvesting. Thermo-responsive coatings, layered on the traditional TCPS surfaces, allowed for the culturing of human-derived stromal cells with similar kinetics observed in the case of uncoated petri dishes. After reaching the confluence, cells could be harvested as contiguous sheets by simply reducing the temperature to 25 °C, with no use of proteolytic enzymes and thus preserving the membrane proteins from digestion. In conclusion to this mainly experimental work, an analysis of the production costs for the developed material is carried out, in order to predict the price for their eventual commercialization. This is an important step to verify if such materials could be competitive on the biomedical market.

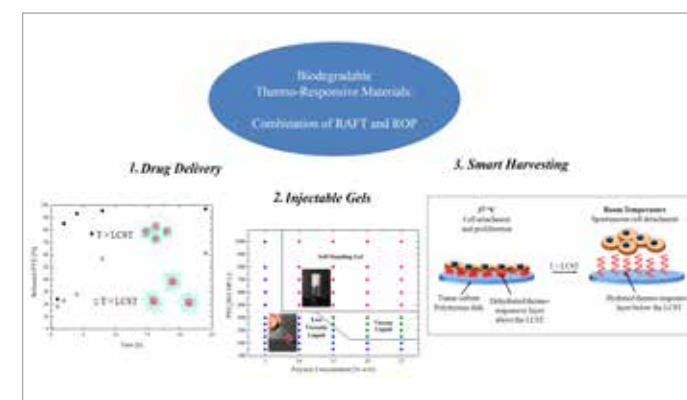


Fig. 2 - Applications of biodegradable thermo-responsive materials explored in this work.

NO_x CONTROL STRATEGIES FOR EXHAUST AFTER-TREATMENT SYSTEMS ON LEAN-BURN VEHICLES

Roberta Villamaina - Supervisor: Enrico Tronconi

The NH₃/urea Selective Catalytic Reduction (NH₃/urea SCR) represents the most effective technology to meet the current limitations regarding the real driving NO_x emissions from lean-burn vehicles.

Nitrogen oxides present in the flue gases are efficiently reduced to harmless N₂ and H₂O through the injection of NH₃/urea over either iron- or copper-exchanged zeolite catalysts coated on monolithic substrates or Vanadia-based catalysts according to the three main reactions of the process (Standard SCR, Fast SCR and NO₂-SCR). However, it presents few points which have to be better understood and improved from a practical point of view. Moreover, the use of only this solution for an even higher overall deNO_x efficiency is no more satisfying and not in line with the upcoming stricter regulations. Due to this, the present PhD work wants to address the debated aspects regarding the NH₃-SCR device as well as a newer strategy which helps in lowering the NO_x emissions.

Despite the NH₃-SCR is a well-established application, the choice of the NH₃-SCR catalyst is a crucial point to enhance the NO_x control. One of the main issues related to the catalytic

systems above mentioned is related to the very poor efficiency at low temperatures. The use of metal promoted zeolite catalysts instead of V-based systems in the automotive field has allowed to improve the NO_x conversions to higher values in the low-temperature region, above all over Cu-zeolite catalysts.

Cu-zeolite catalysts are objects of numerous studies from several research groups. The understanding of the correlation between the catalyst formulation and the presence of a Cu ions population with a specific activity towards the NO_x reduction is being currently discussed and could be useful to improve the efficiency of these catalysts. In this work we investigate the effects of two main catalyst formulation parameters, namely the SiO₂/Al₂O₃ ratio and the Cu loading, on some fundamental aspects of Cu-CHA catalysts for the NH₃-SCR process, such as the interaction between NH₃ and nitrate species with the catalyst surface and the Cu ions reducibility, which could affect the SCR activity. Transient tests were used also to identify the existence of two main Cu cations in the Cu-CHA catalysts and to study how their relative fraction is strictly linked to the SAR and Cu loading (or more generally to the Cu:Al ratio). Several

groups already demonstrated the presence of two of Cu^{II} ions which populate the CHA cages (Z₂Cu^{II} doubly coordinated with two Al atoms, and ZCu^{II}OH, singly coordinated with the zeolite framework) through different advanced techniques (namely DFT calculations, XRD, XAS and XES, FTIR/DRIFT). However, our purpose is to probe experimentally the speciation through simple catalyst characterization tests, such as NH₃ and NO₂ adsorption + TPD or NO+NH₃-TPR runs) over model Cu-CHA catalysts characterized by increasing SAR values and Cu loadings. Moreover, through activity tests under the relevant SCR conditions (Standard SCR, Fast SCR, NO₂-SCR) we tried to correlate the effects of SAR and Cu loading, and in turn the Cu speciation, with the deNO_x activity of Cu-CHA catalysts.

Since the NO_x conversion to harmless gases occurs only at temperatures higher than 170°C (T in which urea decomposition happens) and with efficiency sufficiently high at temperature above 200°C, the adoption of an additional system, which avoids the NO_x emissions at low temperatures, for example during the so-called cold start transient, is required. Currently, new systems devoted to the storage of NOx are being developed and studied, the

so-called Passive NO_x Adsorbers (PNA). These systems are not able to reduce NO_x to N₂ but can only store nitrogen oxides at low temperature and release them at a higher temperature, at which the downstream SCR is active. The most promising formulation was proposed in 2016 by Johnson Matthey and is based on Palladium zeolites. In collaboration with JM we have been working on the study and the characterization of this system in order to evaluate the effect of several process variables, such as the adsorption temperature, the adsorption time, the composition of the gaseous mixture (presence of H₂O, CO, NO₂), in view of optimizing the NO_x storage process and in order to better clarify the mechanism behind this novel technology. In the exhausts after-treatment system of lean-burn engines, the SCR unit is placed downstream of other devices, which help in the removal of other pollutants, such as unburnt hydrocarbons, particulate matter and CO. Like for the SCR, also these catalytic steps can be negatively affected

by phases in which the exhaust temperature is too low to reach a sufficient removal efficiency. The slip of hydrocarbons from its converter units (Diesel Oxidation Catalyst - DOC - or/ and Methane Oxidation Catalyst - MOC) could be also detrimental for the SCR application. As mentioned before, the state-of-the-art catalysts for SCR are based on metal exchanged zeolites: these materials are able to adsorb hydrocarbons, according to their characteristic pore size and acidity, resulting in turn in a competition with NH₃ adsorption or in a block of the active sites or pores upon the formation of intermediate species or coke. On the other hand, they can also take part of the NOx removal, being reducing agents like ammonia (Hydrocarbon SCR). Consequently, the performance of the SCR process needs to be evaluated taking into account also the presence of these species. If the effect of hydrocarbons with medium-long chain is well established, very limited specific information is available in the

case of a CH₄ slip and on how it can interact with the deNO_x process. A possible scenario in which the HC slip is mainly composed of methane is relative to vehicles fuelled with natural gas or dual fuel (Diesel and natural gas) with engines operating in lean conditions. CH₄ emission control is very challenging, being methane a molecule hard to oxidize over its dedicated catalyst (MOC) with high efficiency below 400-450°C, causing a certain slip which could be problematic for the other catalytic after-treatment technologies and, above all, because it has a green-house power much higher than CO₂. Therefore, to complete the picture of the HC slip effect, Fe- and a Cu-exchanged zeolite catalysts, and their combined arrangements, were tested firstly under the typical operating conditions of a Diesel vehicles and then under the typical operating conditions of a lean-burn natural gas in which the methane slip is present.

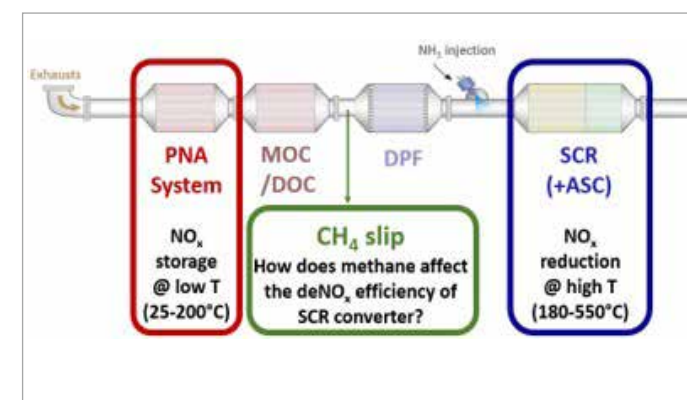


Fig. 1 - The NO_x emission control strategies and issues in an exhaust after-treatment system for lean-burn vehicles