Publishable summary MagPro²Life

« Advanced **Mag**netic nanoparticles deliver smart **Pro**cesses and **Pro**ducts for **Life** «

Researchers from Denmark, Germany, Ireland, Romania, Spain, Switzerland and UK kicked off the EU *MagPro²Life* project on July 1, 2009. The consortium brings together universities, research institutes, SME and enterprises and is coordinated by Solae. The primary objective of this project is to develop pilot lines to introduce nanotechnology based processes into the value chain of existing industries.

Implementation of functional magnetic particles as adsorbents in the bio-processing industry requires the synergistic interplay of a host of components. The two major challenges for implementation of magnetic (nano) particles and composites in industry are the cost-effective large-scale manufacturing of appropriately functionalized super-paramagnetic particles, and the lack of large-scale process technology to separate these particles from the production streams. For this technology to be accepted in industry, this technology must be sustainable, safe, capable of scaling to high production rates, reliable, robust, and economically competitive to current existing downstream processing techniques. The goal of the *MagPro²Life* project is to address these challenges and to demonstrate the use of functional magnetic (nano)particle separation at pilot-scale for selected feed, food, and pharmaceutical products. The *MagPro²Life* project carefully builds on knowledge gained in the NanoBioMag project, funded by the EU under the FP6 program, through a series of focused objectives: (i) development of scalable magnetic particles production technologies; (ii) development of scalable surface functionalizing technologies; (iii) development of large-scale processing technologies to be used with the adsorbents produced by objectives (i) and (ii); and (iv) integration of the previous three objectives to demonstrate that this technology can be implemented for safe, industrial scale bio-separation.

Biotechnologically derived substances for large scale feed, food and pharma applications represent one of the most important sources of new products due to their precisely controlled structural and functional properties, potential for economic and responsible production and overall broad benefits to society through biocompatibility and sustainability. The costs of producing biomaterials are in many cases dominated by separation processes, which can constitute up to 80% of the total cost of production. Using smart magnetic adsorbent particles to selectively separate the target product out of a complex product mixture like the fermentation broth or bio-feed stock can drastically reduce costs. By using magnetic separation and extraction technologies to separate the magnetic carrier particles, novel processing ways emerge.

While the developments of modern gene technology promise to make an increasing number of sophisticated pharmaceuticals available, their enormous production prices will inevitably place strong pressures upon health care systems, even those in wealthier countries. Higher value proteins derived from natural and recombinant sources represent main target bio-products within this project. The range of prices of various industrial protein products vary astronomically up to several million € per kg. High-value pharmaceutical proteins are only affordable for highly developed countries in the western hemisphere, but even in these privileged countries the high cost of these drugs exerts a strongly negative impact on their social health care system. By reducing the production costs of new pharmaceuticals the deliverables of this project will contribute to solving this dilemma.

The project will likely deliver exploitable findings directly applicable to individual products. Practical exploitation will be undertaken by the industrial partners as the first end users. The knowledge database produced during *MagPro²Life* shall guarantee fast market readiness and fast and easy transfer to products other than those employed in the pilot line demonstrations.
The knowledge gained in the areas of process and equipment development and applications will form a basis for creating new process equipment and new high-value products. Equipment when commercialised will be available to industrial sectors such as pharmaceutical, food, chemical and technical industries. It is planned to sell licences for the patented equipment to biotech manufactures.

In the first 18 months of the project, several distinct manufacturing routes were developed to produce assemblies based on inexpensive super paramagnetic particles combined with biomaterials, functional polymers or self-adapting polymers. Recognition systems are constructed to make the adsorbents selective and/or endow them with multifunctional properties. Biomaterials, representing each of the 3 industries for pilot scale demonstration, have been selected and characterized. Focus in the next period will be the comparative characterization of the particle systems and subsequent scale-up of the most promising ones. To successfully compete with classical production processes it is crucial to provide high efficient process equipment especially regarding the recovery of the magnetic adsorbent particles. In the following are the technologies, the consortium is focussing on: Continuous Magnetic Extraction, Magnetic Field Enhanced Centrifugation, and Magnetic Classification. Lab equipment has been used to model the separation systems and preliminary screening of the particle systems has been performed. Focus has been on the pilot scale design of the separation equipment, while efforts for the next period will be the manufacture and optimization of this equipment.

In the second 18 project months, the bio-systems for production and capturing the target molecules have been studied and optimized in all three areas of interest. The separation equipment was built, optimized and benchmarked. The pilot lines where set-up and are ready to run and generate data for the

risk assessment, the economic studies, Life Cycle Analysis and technology benchmarking. Pro-actively, particular focus was set to Anion Exchanger and Cation Exchanger functionalization of the magnetic beads. Sufficient quantities of particle systems were made available for all three pilot plants to trial and run. Additional bead systems of interest are planned in smaller quantities and will be tested on lab scale. They will be scaled up using SuperPro Designer® software in order to offer the possibility to compare these data with the data from the pilot lines.

In the third and last reporting period, the pilot lines have run and demonstrated the MagPro²Life technology. Full data analysis and modeling made an assessment possible on all the aspects of potential successful exploitation. Scale, particle cost and binding capacity have the most significant impact on production cost. We showed that although at current state of the art, still optimization is needed, it is possible to commercially exploit the technology at competitive product prices, with distinct advantages against immediate competing technologies as packed bed and expanded bed technologies. A full comprehensive confidential report has been issued on the technology as explored in this project. A book on the subject matter will be published (Springer) in second quarter of 2014.

Advanced Magnetic nano-particles deliver smart Processes and Products for Life

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Focus

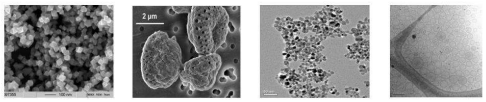
Functional magnetic (nano)particle composites have the potential to enhance the performance and economics of bioseparation processes, because of their extremely high surface areas, rapid binding kinetics, and unique physical and chemical properties. The two major barriers to implement the industrial use of magnetic (nano)particle composites are the safe and effective large-scale manufacturing of appropriately functionalized superparamagnetic particles, and the lack of large-scale process technology to separate these particles from the production streams. The goal of the MagPro²Life project is to address these barriers and demonstrate the use of functional magnetic (nano)particle separation at pilot-scale for selected feed, food, and biopharma products.

Particle synthesis

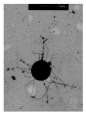
Functionalization - Analytics

In this part the main focus is to develop a set of inexpensive superparamagnetic nano-(composite) materials that have enhanced physical and chemical properties. A large variety of synthesis routes will be established.

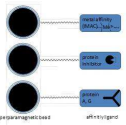
Continuous Suspension Synthesis | Solution / Spray-Process | Laser Pyrolysis/ Precipitation | Rotating Membrane Pore Extrusion



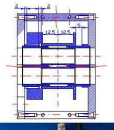
In the next step the magnetic matrices can be coated and biochemically functionalized in many ways to obtain powerful nano-materials with unique adsorptive properties. Techniques are available and developed further to analyze molecular and particle interaction as well as particle detection in the sub-mg per liter scale..



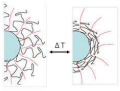
Attachment of phages



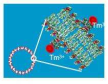
Affinity ligands



Advanced analytical tools: On-line susceptometric magnetic sensor



Thermoresponsive materials

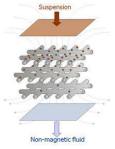


Functionalized vesicles

Process development

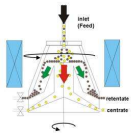
Particle handling

High Gradient Magnetic Filtration



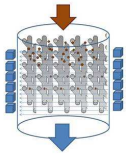
Particles attach to a filter by magnetic forces

Magnetic Field Enhanced Centrifugation



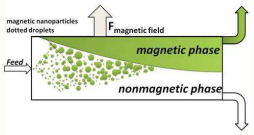
A magnetic filter is cleaned continuously by centrifugation.

Innovative HGMS devices



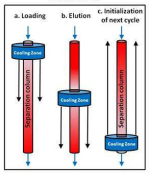
Halbach arrangement as inexpensive alternative to electromagnetic HGMS solutions

Continuous Magnetic Extraction



Nanoparticles are continuously extracted in an aqueous two-phase system supported by an external magnetic field

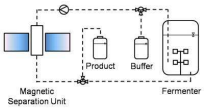
Travelling Cooling Zones Reactor



Combination of a novel separation device with thermoresponsive sorbents

Applications to be tested

In-situ product removal of enzymes applied in Feed during fermentation



- Fermentation intensification via the integration of multi in situ magnetic separation steps
- Prevention of product degradation by capturing e.g. proteases

Recovery of nutraceutical biomolecules from large scale natural Food streams

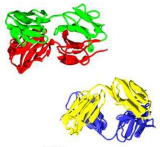
- Exploitation of high value functional food proteins from soy process streams with specific attributes for health, well-being and/or prevention of diseases

Demonstration of the validity of magnetic separation technology for high throughput food industries

Direct capture of high value Biopharma proteins from crude feed streams

- One step purification of novel antibody fragments (FABS) with potential application in therapeutics, medical diagnostics and analytics.

E.Coli Fermentation



FABS

- Systematic benchmarking of magnetic nanoparticle-based bioseparation processes against established techniques like e.g. chromatography

Project partners

Solae	Merck KG	Swiss Federal Institute of Technology
Karlsruhe Institute of Technology	Universidad de Salamanca	Technical University of Denmark
University Birmingham	fzmb GmbH	Romanian Academy- Timisoara Branch
Technische Universität Bergakademie Freiberg	University College Dublin	Bühler AG
National Institute for Laser, Plasma and Radiation Physics	KMPT AG	NIIMT – Cluj Napoca

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