The BIOCHEM project will provide a toolbox which will focus on the needs of SMEs to overcome these barriers and to stimulate demand driven bio-based business in the chemical sector.

The toolbox will include

- bio-based market information and assessment tools for SMEs, including life-cycle methodologies and sustainable business strategy development;
- a European on-line partnering and open innovation resources system;
- access to finance, business planning and fundraising support.

It will then be delivered to selected SMEs through national innovation agencies in the Member states. Each SME will receive a bespoke development plan and personal assistance in the most critical areas.

BIOCHEM is strongly supported by SusChem and reflects the change in emphasis towards supporting innovation as well as research.

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Environmental Biotechnology

[E.1]

Biomass-derived clean biofuels: Techno-economic imperatives

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Keywords: Biofuel products; Pollution abatement; Biomanufacturing; Bimass conversions

"Prevention is better than cure". "Wastes are misplaced resources". Nowadays industrial and environmental technologies are involved in a love-hate relationship which is intensified by the geo-politics of global climate change. This invited keynote talk will examine the cause-and-effect scenarios of current developments and future trends in related biotechnologies under discussion. In particular, bioprocessing insights relevant to techno-economic imperatives of biomass-derived clean biofuels will be presented. Re-visiting the unfinished business on potential bioconversion of lignocellulosic biomass reveals continuing basic scientific hurdles. Within this eco-tapestry there are many intertwined challenges. The overview will evaluate various stages of the production chain: biomass pretreatment, fermentation, strain development, and product recovery. Given their abundance, biodegradable biomass materials impact significantly the global ecology. Agri-food manures and other biomass residues cause widespread environmental pollution in many countries, but these materials are potential renewable resources for biorefineries to produce clean biofuels in contrast with polluting petroleum derivatives. The inherent recalcitrance of cellulosic biomass for bioconversions poses a major challenge; however, recent reduction in the production cost of relevant saccharification enzymes has a major beneficial impact on the manufacturing potentials. Among the current interest, ethanol is the favorite product but biodiesel, biobutanol, methane and hydrogen should not be ignored. The low conversions in the current technologies and the difficulty of safe storage and transportation of biofuel products are additional complications for the marketplace. Alternatives of algae, jatropha and other biological oils also have severe techno-economic constraints. An analysis of the techno-economic parameters indicates conflicting views. The bestavailable-technologies at present have opposite economic realities for developed relative to developing countries which require time for adequate socio-economic development. Despite international geopolitics, challenges at the technology level must be addressed.

[E.2]

The energy crisis-how can microorganisms be put to work?

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Microorganisms are micro-scale factories that can be manipulated to help solve the energy crisis. The ability to obtain a biodiversity of unique genes whose products can act in extreme environment, together with powerful genetic engineering tools, can be utilized to construct efficient biochemical machines, tailormade for the needs of the growing biofuel industry.

The uses of microorganisms include:

- 1. Production of "microdiesel", which unlike regular biofuel, is produced without toxic chemicals and can be made using waste (plant waste or other types of waste) instead of plant oils.
- 2. Converting waste materials-such as lignocellulose-to substrates for biofuel production
- 3. Detoxifying waste material in biodiesel production
- 4. Generate energy as electrical current bacteria are capable of using anything from decaying plant and animal matter to toxic organic pollutants such as benzene to produce electricity.

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[E.3]

Bioelectrochemical dechlorination of trichloroethene: From electron transfer mechanisms to process scale-up

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² Water Research Institute, National Research Council, Italy Keywords: Bioelectrochemical systems; Reductive dechlorination; TCE; electron transfer

Current in situ bioremediation approaches for chlorinated solvents in groundwater typically involve the stimulation of microbial reductive dechlorination (RD) processes through the subsurface injection of organic electron donors to provide native dechlorinating populations with the electrons required for contaminants reduction.

Recently, bioelectrochemical systems (BES), in which solid-state (e.g., graphite-based) electrodes are employed as direct electron donors (in place of organic electron donors) in the RD of chlorinated solvents, have been proposed. Main advantages of the bioelectrochemical approach include a greater control over electron delivery and consumption and a no longer required injection of chemicals into the subsurface.

Investigations conducted at our laboratories have shown that bioelectrochemical stimulation of RD can be achieved via at least two different electron transfer mechanisms: (1) the electrolytic generation of molecular hydrogen, which in turn serves as an electron donor for the RD; (2) the direct exchange of electrons between the electrode and the dechlorinating bacteria, growing attached at the electrode surface in the form of an "electro-active" biofilm. The latter can also be enhanced by the presence of redox mediators immobilized at the electrode surface. The relative contribution of these different mechanisms depends on several factors such as the working electrode (i.e., cathode) potential, the electrode surface area, and biomass density. Since the predominance of one electron transfer mechanism over the other has direct and remarkable implications on the kinetics, efficiency, and selectivity of the dechlorination process, a careful optimization of the above describe factors is needed.

The study aimed to evaluate the implications related to the scaling-up of a bioelectrochemical process for TCE-dechlorination. For this purpose, the performance of different bioelectrochemical systems, operated at low or high (electrode) surface area to reactor volume ratios was analyzed, with main reference to involved electron transfer mechanisms, dechlorination kinetics, and electron capture efficiencies.

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[E.4]

Combined electrochemical and biological treatment for tetracycline and tylosin removal

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Keywords: Combined process; Electrochemical pre-treatment; Biological treatment; Tetracycline; Tylosin

This work aims to study the degradation of tetracycline and tylosin by an integrated process including electrochemical and biological ways.

Tetracycline and tylosin antibiotics, widely used antibacterial in the world, are of a special concern because of their extensive use in human and veterinary medicine. Approximatively 70% of the applied drugs are not metabolized or absorbed in the body. The presence of tetracycline residues and their potential to promote growth of resistant bacteria pose adverse health effects to humans.

Microbial toxicity biotests showed that at a relatively high concentration, tetracycline and tylosin are not biodegradable and have inhibitory action. So, their direct degradation by conventional biological way is not appropriate. However, antibiotics could be fragmented by electrochemical treatment which seems to be an attractive way to destroy recalcitrant organic contaminants such as antibiotics.

Cyclic voltammetry with a vitreous carbon electrode revealed a good electrochemical activity for both antibiotics. Such behavior led us to consider an electrochemical pre-treatment in order to decrease the toxicity and improve the biodegradability of studied antibiotics.

Electrochemical treatment was performed in a flow cell. The effect of operating conditions such as concentration, flow percolation and electrical current density was investigated. Tetracycline and tylosin were reduced and/or oxidized in neutral and acidic media. More than 90% of tetracycline was transformed.

High performance liquid chromatography (HPLC), UV–visible (UV–vis) spectroscopy and total organic carbon (TOC) analysis revealed that although a complete disappearance of tetracycline and tylosin, the level of mineralization remained low. Biological treatment of the electrolysed solutions was performed in batch with activated sludge taken from a local municipal wastewater treatment plant, and the evolution of the biological treatment was monitored through BOD₅ and COD analysis.

[E.5]

Effect of aerobic stabilization on biomass activity

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The study was aimed to decrease the organic matter and the biomass content of the sludge taken from the aeration unit of a municipal wastewater treatment plant through aerobic stabilization. The efficiency of the stabilization was assessed monitoring pH, suspended solids (SS), volatile suspended solids (VSS), total and dissolved organic carbon (TOC, DOC), nitrate, nitrite, phosphate, and heavy metals parameters. The oxygen uptake rate (OUR) measurements were conducted to determine active biomass concentration. The results of the OUR assays were modeled using Activated Sludge Model No:3 (ASM3). DAPI staining targeting the genomic DNA of the cells were used to count total cells. Amount of the total active bacteria were determined by an oligonucleotide probe targeting rRNA content of the cells. On the 30th day of the stabilization the SS, VSS and TOC removal was 22%, 28% and 55%, respectively. Based on respirometric results, the ratios of the active biomass were decreased to 35% and 21% for the 17th and 30th day of the stabilization, respectively. According to the active bacteria quantification, the active cells ratio decrease down to the 22% at the 30th day of the stabilization. After 15 days of the stabilization, the endogenous respiration phase was started due to the complete consumption of the extracellular organic carbon, and eventually a significant decrease in the amount of the total cells and total active bacteria was achieved. It is noteworthy that the respirometric results were in accordance with the molecular analyses results which prove that both methods can be carried out to quantify the amount of the active biomass. Such results have significant implications relative to the activity decrease quantification of the biomass as well as its further application potentials after aerobic sludge stabilization.



Fig. 1. The images of cells stained with DAPI (a) raw sludge (b) after 30 days aerobic stabilization, and hybridized with epifluorescent EUBmix probe (c) raw sludge (d) after 30 days aerobic stabilization.

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