



ENVIS CENTER on ENVIRONMENTAL BIOTECHNOLOGY

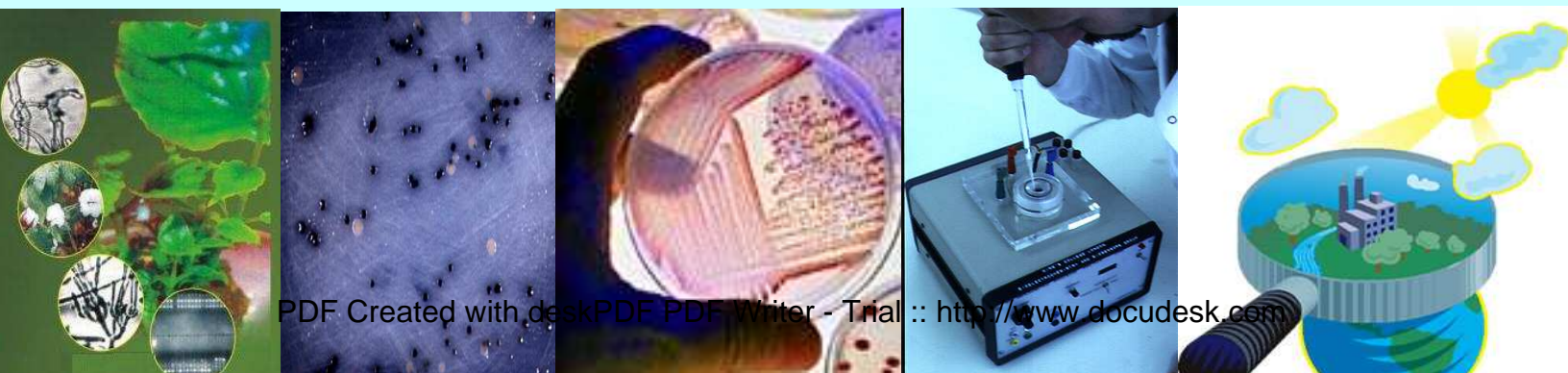
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ENVIS CENTRE

on

ENVIRONMENTAL BIOTECHNOLOGY

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BACKGROUND

Environmental Information System (ENVIS) is established in the year 1984 as a network of Information Centres. It is planned by the Ministry of Environment and Forest. Aim of this centre is to provide descriptive and environmental subject related numerical data. Now 78 centres are working under this network on various subject areas in the country. The focal point of this network is situated at the Ministry of Environment and Forest, Government of India, New Delhi.

This ENVIS Centre is established for studies on Environmental Biotechnology at the Department of Environmental Science, University of Kalyani, Nadia-741235, West Bengal.

The objective of this centre is to collect data related to the above mentioned subject, from different major libraries mainly in West Bengal and also from other states in India, through consultation with different journals, Annual Reviews, Internet and to generate a database and create a website uploaded with these information. Besides, we publish biannually Abstract Volume on our thematic area Environmental Biotechnology under fourteen subheads. The volume contains abstracts of scientific articles from relevant national and international journals. Viewpoint of this abstract volume is to help the interested research workers, scientists, administrators and the general people.

This is the 14th publication of Abstract Volume of this ENVIS Centre. This contains the abstracts of research papers collected from the various areas of Environmental Biotechnology from different journals published in June, 2009. In this issue, various topics like Bioenergy, Bioengineering, Bio-degradation, Bio-remediation, Bio-transformation etc. have been covered. We are grateful to the various libraries and their staff for their cooperation extended to us during the collection of the articles.

Abstract Format

The format of the abstract is as follows:

Abstract : The abstracts are arranged in different subheads.

Author: Name of the authors are given in the order in which they appear in the original document. These names are given in succession.

Address of Authors: Address of the author is given in parenthesis at the end of the authors name. When the address of any other author is found, it is written after wards delimited by stop(.).

Locus: The name of the journal is followed by the volume number, issue number, year of publication and the page no.

GENERAL INFORMATION

Abstract have been taken directly from source documents like research report, journals, internet, seminar proceedings, standards and patents. All the resources are published within last six months.

Abstract are broadly classified and arranged under the following 14 heads:

Bioaccumulation: Bioaccumulation means an increase in the concentration of a chemical in a biological organism over time, compared to the chemical's concentration in the environment. Compounds accumulate in living things whenever they are taken up and stored at a rate faster than they are broken down (metabolized) or excreted. Understanding the dynamic process of bioaccumulation is very important in protecting human beings and other organisms from the adverse effects of chemical exposure, and it has become a critical consideration in the regulation of chemicals.

Bioremediation: It is a clean-up technology that uses naturally occurring microorganisms to degrade hazardous substances into less toxic or nontoxic compounds. The microorganisms may:

1. Ingest and degrade organic substances as their food and energy source,
2. Degrade organic substances, such as chlorinated solvents or petroleum products, that are hazardous to living organisms, including humans, and degrade the organic contaminants into inert products.

As the microorganisms occur naturally in the environment they are likely to pose little risks of contamination.

Bio-Transformation: This is a process of Biological changes of complex compounds to simpler one or toxic to non-toxic and vice-versa. Several microorganisms are capable of transforming a variety of compounds found in nature but generally in case of synthetic compounds they are unable to show any appropriate action. Biotransfer appears to be one of the major detoxication methods known so far.

Biomarker: It is a biological response to a chemical that gives a measurement of exposure and, sometimes, of toxic effect. It can be defined as any kind of molecule which indicate the existence (past or present) of living organisms. In particular, in the fields of geology and astrobiology biomarkers are also known as biosignatures. However, in environmental science a bio-markers can also be used to indicate exposure to various environmental substances in epidemiology and toxicology.

Biofertilizer: To reduce the impact of excess chemical fertilizers in the field of agriculture the biofertilizer is being considered as a potential tool; biologically fixed nitrogen

is such a source which can supply an adequate amount of Nitrogen to plants and other nutrients to some extent. Many free living and symbiotic bacteria, which fix atmospheric Nitrogen are used as biofertiliser material as a substitute for Nitrogen fertilizer. In general two types of biofertiliser are used

1. Bacterial Biofertilizer
2. Algal Biofertilizer

Biocomposting: It involves combining organic materials under conditions that enables them to decompose more quickly than they would in nature. Think about logs and leaves on the ground in a forest. The leaves will break down and disappear within a year. Logs of course will take much longer to crumble away. Composting is the process of converting all biodegradable wastes into organic manure. In composting process certain input should be made into waste to convert the process in a short time.

Biopesticide: Pest control by biological antagonism appears to be very useful tool in recent years. Bacterial pesticides are being developed. *Heliothis* complex, which lives in close association with plant roots, consists of two major crop pests budworm and ball worm. Biological insecticides against both these insects are being prepared by transfer of a gene from *Bacillus thuringiensis*

Biodegradation: It is nature's way of recycling wastes, breaking down organic matter into nutrients that can be used by other organisms. "Degradation" means decay, and the "bio-" prefix means that the decay is carried out by a huge assortment of bacteria, fungi, maggots, worms, and other organisms that eat dead material and recycle it into new forms.

In the nature, nothing is known as waste, because everything gets recycled. The waste products from one organism become the food for others, providing nutrients and energy while breaking down the waste organic matter. Some organic materials may break down much faster than others, but all will eventually decay.

By harnessing these natural forces of biodegradation, people can reduce wastes and clean up some types of environmental contaminants. Through **composting**, we accelerate natural biodegradation and convert organic wastes to a valuable resource.

Biosensor: Biosensor represents biophysical devices, which can detect the presence and measure the quantities of specific substances in a variety of environments. These specific substances may include sugars, proteins, or humas and variety of toxins in the industrial effluents. In designing a biosensor an enzyme or an antibody or even microbial cells are associated with microchip devices, which are used for quantitative estimate of a substance.

Bioengineering: It is a developing speciality featuring a multidisciplinary approach to the solution of problems in medicine and biology, based on the application of

advances in science, engineering and technology. It generally engineers the biological processes through biotechnological or genetic engineering interventions. It may also be a broad-based engineering discipline that involve product design, sustainability and analysis of biological systems.

Pollen-Biotechnology: This is a new field of science dealing with the pollen chemistry and allergenicity of aerospora. This subject also covers genetic manipulation of pollen development of haploid culture. Such haploid plants have immense values in genetic research.

Biotechnology Policy Issue: Biotechnology appears to be an emerging science in present decades. Genetic manipulation and development of genetically modified organism in human welfare is now showed a potential prospect and risk. Thus, researches and application of Biotechnology in diverse field is a major policy issue in the present decades.

Agricultural Biotechnology: Over the years, tremendous success has been made in diverse field of agriculture by applying Biotechnology. It includes development of genetically modified crops, genetic improvement in sericulture practices, improvement in Biofertilizer development and similar other aspects. Production of pest and disease resistant crop is also being considered to be an emerging area of Agricultural Biotechnology.

Bioenergy: In recent decades, efforts have been made for evolving were non-polluting bioenergy sources or energy generation from organic wastes and biomass. These are all ecofriendly solutions. Biomass energy supply-demand balances have become a component of energy sector analysis and planning and is propelled huge importance in the countries. Biomass, Biogas, Hydrogen are the example of Bioenergy.

ABBREVIATIONS USED IN ADDRESSES AND CITED JOURNALS

Acad	Academy	Chem	Chemistry
Adm	Administration	Cheml	Chemical
Admn	Administrative	Clinl	Clinical
Adv	Advance	Co	Company
Agri	Agriculture	Coll	College
Agricl	Agricultural	Comm	Committee
Amer	American	Commn	Commission
An	Annual	Comp	Comparative
Analyt	Analytical	Conf	Conference
Anat	Anatomy	Conv	Convention
Anim	Animal	Conserv	Conservation
Ann	Annals	Contl	Control
Appl	Applied	Contam	Contamination
Arch	Archives	Corp	Corporation
Archaeo	Archaeology	Coun	Council
Archaeol	Archaeological	Cult	Culture
Architect	Architecture	Cultl	Cultural
Assoc	Association	Curr	Current
Asst	Assistant	Dept	Department
Atom	Atomic	Dev	Development
Bacterio	Bacteriology	Develop	Developmental
Bacteriol	Bacteriological	Dig	Digest
Bd	Board	Div	Division
Bio	Biology	Divl	Divisional
Biochem	Biochemistry	Dte	Directorate
Biocheml	Biochemical	Dy	Deputy
Bioengg	Bioengineering	Eco	Ecology
Biol	Biological	Ecol	Ecological
Biometeo	Biometeorology	Econ	Economics
Biophys	Biophysics	Ecosys	Ecosystem
Biometeol	Biometeorological	Ecotoxicol	Ecotoxicology
Biotech	Biotechnology(s)	Endocrinol	Endocrinological
Biotechno	Biotechnology	Engg	Engineering
Biotechnol	Biotechnological	Engrs	Engineers
Bldg	Building	Env	Environment
Bot	Botany	Environ	Environmental
Botl	Botanical	Epidemic	Epidemiology
Br	Branch	Epidemiol	Epidemiological
Bull	Bulletin	Estd	Establishment
Cent	Centre	Ethnopharmacol	Ethnopharmacology
Centl	Central	Expt	Experiment

Exptl	Experimental	Microbiol	Microbiological
Fac	Faculty	Min	Ministry
Fd	Food	Monit	Monitoring
Fedn	Federation	Myco	Mycology
Fert	Fertiliser	Mycol	Mycological
Fmg	Farming	Nat	Natural
Gaz	Gazette	Natl	National
Genet	Genetics	N-E	North Eastern
Geo	Geology	Nut	Nutrition
Geogr	Geography	No	Number
Geogr1	Geographical	Occ	Occasional
Geol	Geological	Occupl	Occupational
Geosci	Geoscience	Oceanogr	Oceanography
Govt	Government	Org	Original
Hist	History	Orgc	Organic
Hlth	Health	Orgn	Organisation
Hort	Horticulture	Pharmaco	Pharmacology
Hosp	Hospital	Pharmacol	Pharmacological
Hydro	Hydrology	Phyl	Physical
Hydrol	Hydrological	Patho	Pathology
Immuno	Immunology	Pathol	Pathological
Immunol	Immunological	Petrochem	Petrochemical
Ind	Industry	Petro	Petrology
Inf	Information	PG	Post Graduate
Inst	Institute	Phys	Physics
Instn	Institution	Physio	Physiology
Int	International	Phytopath	Phytopathology
Irrig	Irrigation	Phytopathol	Phytopathological
J	Journal	Plang	Planning
Lab	Laboratory	Polln	Pollution
Lett	Letter(s)	Proc	Proceedings
Ltd	Limited	Prot	Protection
Malario	Malariology	Pub	Publication
Malariol	Malariological	Pvt	Private
Manag	Management	Qlty	Quality
Med	Medicine	Qr	Quarter
Medl	Medical	Rad	Radiation
Metab	Metabolism	Radio	Radiology
Metall	Metallurgy	Radiol	Radiological
Metallurg	Metallurgical	Rd	Road
Meteo	Meteorology	Recd	Received
Meteol	Meteorological	Reg	Region
Microbio	Microbiology	Regl	Regional

Rep	Report	Surv	Survey
Reptr	Reporter	Syst	System
Res	Research	Tax	Taxonomy
Rev	Review	Techl	Technical
Sch	School(s)	Techno	Technology
Sci	Sciences(s)	Technol	Technological
Scient	Scientific	Toxico	Toxicology
S-E	South East	Toxicol	Toxicological
Sec	Section	Transc	Transcations
Sect	Sector	Transpt	Transportation
Semin	Seminar	Trng	Training
Ser	Services	Trop	Tropical
Soc	Society	Univ	University
Socl	Social	Util	Utilisation
Stat	Statistics	Vet	Veterinary
Statl	Statistical	Zoo	Zoology
Stnd	Standard(s)	Zool	Zoological
Stud	Study/ (eis)		

Bioaccumulation

Valeska Contardo-Jara^{a, 1}, Eva Klingelmann^{b, 1}, and Claudia Wiegand^{a, c}. (^aLeibniz-Institute of Freshwater Ecology and Inland Fisheries, Department of Inland Fisheries, Biochemical Regulation, Müggelseedamm 301, 12587 Berlin, Germany, ^bTechnische Universität Berlin/Berlin Institute of Technology, Department of Ecology, Chair of Soil Protection, Salzufer 12, 10587 Berlin, Germany, ^cHumboldt University Berlin, Faculty of Biology, Unter den Linden 6, 10099 Berlin, Germany). **Bioaccumulation of glyphosate and its formulation Roundup Ultra in *Lumbriculus variegatus* and its effects on biotransformation and antioxidant enzymes. Environmental Pollution, Volume 157(1) (2009): 57-63**

The bioaccumulation potential of glyphosate and the formulation Roundup Ultra, as well as possible effects on biotransformation and antioxidant enzymes in *Lumbriculus variegatus* were compared by four days exposure to concentrations between 0.05 and 5 mg L⁻¹ pure glyphosate and its formulation. Bioaccumulation was determined using ¹⁴C labeled glyphosate. The bioaccumulation factor (BCF) varied between 1.4 and 5.9 for the different concentrations, and was higher than estimated from log *P*_{ow}. Glyphosate and its surfactant POEA caused elevation of biotransformation enzyme soluble glutathione *S*-transferase at non-toxic concentrations. Membrane bound glutathione *S*-transferase activity was significantly elevated in Roundup Ultra exposed worms, compared to treatment with equal glyphosate concentrations, but did not significantly differ from the control. Antioxidant enzyme superoxide dismutase was significantly increased by glyphosate but in particular by Roundup Ultra exposure indicating oxidative stress. The results show that the formulation Roundup Ultra is of more ecotoxicological relevance than the glyphosate itself.

Roundup Ultra is of more ecotoxicological relevance than the active ingredient, glyphosate, to *Lumbriculus variegatus* regarding accumulation potential and enzymatic responses.

Keywords: Glyphosate; Roundup Ultra; Bioaccumulation; Biotransformation; Oxidative stress

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The spatial distribution of an important air pollutant class, three-ring polycyclic aromatic hydrocarbons and their derivatives (PAH-3), has been monitored for the Greater Cologne Conurbation (GCC) using pine needle as passive samplers. The GCC comprises one of the most heavily populated, trafficked, and industrialized regions in Germany. Here, 71 locations covering 3600 km² were sampled and, for the first time, isopleths maps constructed to investigate the regional variability in PAH-3 concentration and composition. The highest PAH-3 loads on needles (1000–1500 ng g⁻¹) were detected downwind of three lignite fuelled power plants, followed by Cologne City (600–700 ng g⁻¹) and smaller towns (400–600 ng g⁻¹), whereas rural

and forest regions yielded PAH-3 loads of 60–300 ng g⁻¹. PAH-3 ratios facilitated source reconciliation, with high dibenzothiophene versus retene values indicating lignite combustion and high 9/(9 + 1)-methylphenanthrene ratios depicting traffic emissions in inner cities. PAH-3 ratios depended on topography and outlined the heavily industrialized Rhine Valley, demonstrating atmospheric dispersal of PAH-3.

Regional high-resolution biomonitoring identified lignite combustion in power plants to dominate over urban traffic and other emission sources.

Keywords: Spatial mapping; Methylated phenanthrenes; Dibenzothiophene; Cyclopenta[def]phenanthrene; Source discrimination

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We examined the effect of ozone (O₃) on Norway spruce (*Picea abies*) needle epicuticular wax over three seasons at the Kranzberg Ozone Fumigation Experiment. Exposure to 2× ambient O₃ ranged from 64.5 to 74.2 μl O₃ l⁻¹ h AOT40, and 117.1 to 123.2 nl O₃ l⁻¹ 4th highest daily maximum 8-h average O₃ concentration. The proportion of current-year needle surface covered by wax tubes, tube aggregates, and plates decreased (*P* = 0.011) under 2× O₃. Epistomatal chambers had increased deposits of amorphous wax. Proportion of secondary alcohols varied due to year (*P* = 0.004) and O₃ treatment (*P* = 0.029). Secondary alcohols were reduced by 9.1% under 2× O₃. Exposure to 2× O₃ increased (*P* = 0.037) proportions of fatty acids by 29%. Opposing trends in secondary alcohols and fatty acids indicate a direct action of O₃ on wax biosynthesis. These results demonstrate O₃-induced changes in biologically important needle surface characteristics of 50-year-old field-grown trees.

Free-air ozone exposure induced changes in needle wax characteristics of mature *Picea abies*.

Keywords: Epicuticular wax; Free-air exposure; Mature trees; Norway spruce (*Picea abies*); Ozone

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of Nature Management, Siberian Branch of Russian Academy of Sciences, Ulan-Ude, Buryatia 670047, Russia). Accumulation features and temporal trends of PCDDs, PCDFs and PCBs in Baikal seals (*Pusa sibirica*). *Environmental Pollution*, Volume 157(3) (2009): 737-747

This study investigated the accumulation features and temporal trends of PCDD/Fs, dioxin-like PCBs (DL-PCBs) and non-dioxin-like PCBs (NDL-PCBs) in the blubber of Baikal seals collected in 1992 and 2005. DL-PCBs (480–3600 ng/g) and NDL-PCBs (980–35,000 ng/g) were dominant contaminants. Concentrations of PCDDs and PCBs in males were significantly higher than in females. In males, age-dependent accumulation was observed for PCDDs, mono-*ortho* PCBs and NDL-PCBs. PCDFs and non-*ortho* PCBs showed no such trends, implying that exposure of seals to these contaminants has been decreasing in recent years. No decreasing temporal trend was observed for PCDDs, mono-*ortho* PCBs and NDL-PCBs, suggesting that Baikal seals are still exposed to PCDDs and PCBs. TEQs of PCDDs and mono-*ortho* PCBs in seals collected in 2005 accounted for 62–77% of total TEQs. The TEQ levels in 40% of the specimens exceeded the threshold level for immunosuppression observed in harbor seals (209 pg/g). Concentrations of PCDDs and PCBs remain high in Baikal seals.

Keywords: PCDD/Fs; PCBs; Baikal seals (*Pusa sibirica*); Temporal trends

Gui-Lan Niu^{a, b}, Jun-Jie Zhang^a, Shuo Zhao^{a, b}, Hong Liu^a, Nico Boon^c and Ning-Yi Zhou^a (^aState Key Laboratory of Virology, Wuhan Institute of Virology, Chinese Academy of Sciences, Wuhan 430071, China, ^bGraduate School, Chinese Academy of Sciences, Beijing 100049, China, ^cLaboratory of Microbial Ecology and Technology (LabMET), Ghent University, Coupure Links 653, B-9000 Gent, Belgium). Bioaugmentation of a 4-chloronitrobenzene contaminated soil with *Pseudomonas putida* ZWL73. *Environmental Pollution*, Volume 157(3) (2009): 763-771

The strain *Pseudomonas putida* ZWL73, which metabolizes 4-chloronitrobenzene (4CNB) by a partial-reductive pathway, was inoculated into lab-scale 4CNB-contaminated soil for bioaugmentation purposes in this study. The degradation of 4CNB was clearly stimulated, as indicated with the gradual accumulation of ammonium and chloride. Simultaneously, the diversity and quantity of cultivable heterotrophic bacteria decreased due to 4CNB contamination, while the quantity of 4CNB-resistant bacteria increased. During the bioaugmentation, denaturing gradient gel electrophoresis analysis showed the changes of diversity in dominant populations of intrinsic soil microbiota. The results showed that *Alphaproteobacteria* and *Betaproteobacteria* were not distinctly affected, but *Actinobacteria* were apparently stimulated. In addition, an interesting dynamic within *Acidobacteria* was observed, as well as an influence on ammonia-oxidizing bacteria population. These combined findings demonstrate that the removal of 4CNB in soils by inoculating strain ZWL73 is feasible, and that specific populations in soils rapidly changed in response to 4CNB contamination and subsequent bioaugmentation.

Pseudomonas putida ZWL73 can accelerate 4CNB removal in lab-scale soils, causing dynamic changes within intrinsic *Actinobacteria* and *Acidobacteria*.

Keywords: 4-Chloronitrobenzene; Ammonia-oxidizing bacteria; Bioaugmentation; Denaturing gradient gel electrophoresis; *Pseudomonas putida* ZWL73

William Hartley^a, Nicholas M. Dickinson^a, Rafael Clemente^b, Christopher French^{a, 1}, Trevor G. Pearce^c, Shaun Sparke^a and Nicholas W. Lepp^a. (^aSchool of Biological and Earth Sciences, Liverpool John Moores University, Byrom Street, Liverpool L3 3AF, UK, ^bDepartment of Soil and Water Conservation and Organic Waste Management, Centro de Edafología y Biología Aplicada del Segura, CSIC, Apartado 4195, 30080 Murcia, Spain, ^cBiological Sciences Division, Lancaster Environment Centre, Lancaster University, Lancaster LA1 4YQ, UK). **Arsenic stability and mobilization in soil at an amenity grassland overlying chemical waste (St. Helens, UK). Environmental Pollution, Volume 157(3) (2009):847-856**

A 6.6 ha grassland, established on a former chemical waste site adjacent to a residential area, contains arsenic (As) in surface soil at concentrations 200 times higher than UK Soil Guideline Values. The site is not recognized as statutory contaminated land, partly on the assumption that mobility of the metalloid presents a negligible threat to human health, groundwater and ecological receptors. Evidence for this is evaluated, based on studies of the effect of organic (green waste compost) and inorganic (iron oxides, lime and phosphate) amendments on As fractionation, mobility, plant uptake and earthworm communities. Arsenic mobility in soil was low but significantly related to dissolved organic matter and phosphate, with immobilization associated with iron oxides. Plant uptake was low and there was little apparent impact on earthworms. The existing vegetation cover reduces re-entrainment of dust-blown particulates and pathways of As exposure via this route. Minimizing risks to receptors requires avoidance of soil exposure, and no compost or phosphate application.

Stabilization of alkali industry waste requires careful management to minimise soil arsenic mobilization and dispersal to the wider environment.

Keywords: Brownfield; Phytoremediation; Arsenic; Soil; Risk assessment

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To evaluate the biomagnification extent of polybrominated diphenyls ethers (PBDEs) and polychlorinated biphenyls (PCBs) in a highly contaminated freshwater food web from South China, trophic magnification factors (TMFs) for 18 PBDE congeners and 53 PCB congeners were calculated. The TMF values ranged 0.26–4.47 for PBDEs and 0.75–5.10 for PCBs. Forty-five of 53 PCBs and BDEs 47, 100 and 154 had TMFs greater than one, suggesting their biomagnification in the present food web. The TMFs for PBDEs were generally smaller than those for PCBs with the same degree of halogenation, indicating a lower biomagnification potential for PBDEs compared to PCBs. For PCBs, it followed a parabolic relationship between TMFs and $\log K_{OW}$ (octanol-water partition coefficient). However, this relationship was not

significant for PBDEs, possibly due to the more complex behaviors of PBDEs in the food web (e.g., metabolism), compared to that of PCBs.

Forty-five of 53 PCBs magnified in the freshwater food web, while only BDEs 47, 100 and 154 significantly magnified in the same food web.

Keywords: PBDEs; PCBs; Trophic magnification factors (TMFs); South China

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Arsenic is known to accumulate with iron plaque on macrophyte roots. Three to four years after the Aznalcóllar mine spill (Spain), residual arsenic contamination left in seasonal wetland habitats has been identified in this form by scanning electron microscopy. Total digestion has determined arsenic concentrations in thoroughly washed 'root + plaque' material in excess of 1000 mg kg⁻¹, and further analysis using X-ray absorption spectroscopy suggests arsenic exists as both arsenate and arsenite. Certain herbivorous species feed on rhizomes and bulbs of macrophytes in a wide range of global environments, and the ecotoxicological impact of consuming arsenic rich iron plaque associated with such food items remains to be quantified. Here, greylag geese which feed on *Scirpus maritimus* rhizome and bulb material in areas affected by the Aznalcóllar spill are shown to have elevated levels of arsenic in their feces, which may originate from arsenic rich iron plaque.

Accumulation of metals with iron plaque on macrophyte roots in wetlands poses an ecotoxicological risk to certain herbivores.

Keywords: Herbivorous wildlife; Metalloids; Food chain transfer; Wetlands; Phytoremediation

R.M. Godinho^a, T.G. Verburg^b, M.C. Freitas^a and H.Th. Wolterbeek^b. (^aInstituto Tecnológico e Nuclear Reactor, Apartado 21, E.N. 10, 2686-953 Sacavém, Portugal, ^bDepartment of Radiation, Radionuclides and Reactors, Section RIH (Radiation and Isotopes in Health), Faculty of Applied Sciences, Technical University Delft, Mekelweg 15, 2629 JB Delft, The Netherlands). **Accumulation of trace elements in the peripheral and central parts of two species of epiphytic lichens transplanted to a polluted site in Portugal. Environmental Pollution, Volume 157(1) (2009): 102-109**

This paper compares the dynamics, i.e. the rates of change in element concentrations of young and older lichen thallus parts, of one foliose and one fruticose lichen, during a transplant experiment to a polluted site. Both lichen parts respond to environmental changes. Here, differential accumulation suggests that differential constitution leads to differential uptake and

release, and/or the overall behaviour is partly due to internal translocation and regulation mechanisms within the whole lichen. For thallus parts, internal translocation should be taken into account as one more factor affecting lichen “memory length”. Young parts of the thallus presented higher rates of change, but different lichen parts accumulate different elements to different extents. Therefore tissue selection in monitoring may depend on the element of interest, and cannot be made into a generalized approach in survey set-ups: the choice depends on the element.

Thallus age and type affect the rate of change of element concentrations in lichens as induced by changes in ambient environmental conditions.

Keywords: *Flavoparmelia caperata*; *Evernia prunastri*; Biomonitoring; Transplant; Age

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The research on the effects of microcystins on aquatic plants has increased. Some aquatic plants have some tolerance to microcystins but the mechanism of the tolerance is still unknown. In this experiment, we used microcystins of different concentrations to study the toxic effect in *Lemna minor* and *Myriophyllum spicatum*. Experiments were carried out with a range of microcystins levels (equivalent to 0, 0.1, 0.5, 1.0, and 4.3 mg/L). The growth of *L. minor* (as fresh weight) and chlorophyll a content were significantly reduced and superoxide dismutase (SOD) activity was significantly decreased at microcystins concentration up to 0.5 mg/L. The growth of *M. spicatum* was affected, only weakly, by microcystins and 0.5 mg/L and these treatments caused significant decrease in chlorophyll a content. Besides, the SOD activity of *M. spicatum* positively correlated to microcystins concentration ($P < 0.01$). The result indicated that *M. spicatum* was more tolerant to microcystins than *L. minor* and the induced SOD activity may contribute to the tolerance. The experiment also indicated that catalase (CAT) activity was not significantly influenced by microcystin for both the two tested aquatic plants.

Keywords: microcystin • *Lemna minor* • *Myriophyllum spicatum* • SOD • CAT

Yan Gong^{1,2}, Lirong Song¹, Xingqiang Wu^{1,2}, Bangding Xiao¹, Tao Fang¹, Jiantong Liu^{1*}. (¹Institute of Hydrobiology, Chinese Academy of Sciences, Wuhan 430072, People's Republic of China, ²Graduate School of the Chinese Academy of Sciences, Beijing 100039, People's Republic of China. *Correspondence to Jiantong Liu, Institute of Hydrobiology, Chinese Academy of Sciences, Wuhan 430072, People's Republic of China). **Effects of arsenate on microcystin content and leakage of *Microcystis* strain PCC7806 under various phosphate regimes. *Environmental Toxicology*, Volume 24(1) (2009): 87 – 94**

Both arsenic pollution and eutrophication are prominent environmental issues when considering the problem of global water pollution. It is important to reveal the effects of arsenic species on cyanobacterial growth and toxin yields to assess ecological risk of arsenic pollution or at least understand naturally occurring blooms. The sensitivity of cyanobacteria to arsenate has often been linked to the structural similarities of arsenate and phosphate. Thus, we approached the

effect of arsenate with concentrations from 10^{-8} to 10^{-4} M on *Microcystis* strain PCC7806 under various phosphate regimes. The present study showed that *Microcystis* strain PCC7806 was arsenate tolerant up to 10^{-4} M. And such tolerance was without reference to both content of intra- and extra-cellular phosphate. It seems that arsenate involved the regulation of microcystin synthesis and cellular polyphosphate contributed to microcystin production of *Microcystis* responding to arsenate, since there was a positive linear correlation of the cellular microcystin quota with the exposure concentration of arsenate when the cells were not preconditioned to phosphate starvation. It is presumed that arsenate could help to actively export microcystins from living *Microcystis* cells when preconditioned to phosphate starvation and incubated with the medium containing $1 \mu\text{M}$ phosphate. This study firstly provided evidence that microcystin content and/or release of *Microcystis* might be impacted by arsenate if it exists in harmful algal blooms.

Keywords: arsenate • phosphate • *Microcystis* • microcystins • cyanobacterium

Pinar Yilmazer ^{*}, Nurdan Saracoglu. (Department of Chemical Engineering, Gazi University, Maltepe, Ankara 06570, Turkey. ^{*}Correspondence to Pinar Yilmazer, Department of Chemical Engineering, Gazi University, Maltepe, Ankara 06570, Turkey). Bioaccumulation and biosorption of copper(II) and chromium(III) from aqueous solutions by *Pichia stipitis* yeast. *Journal of Chemical Technology & Biotechnology*, Volume 84(4) (2009): 604 – 610

BACKGROUND: Bioaccumulation and biosorption by *Pichia stipitis* yeast has not yet been explored. This paper evaluates, for the first time, the use of both viable and nonviable *P. stipitis* yeast to eliminate Cu(II) and Cr(III) from aqueous solutions. The effect of Cu(II) and Cr(III) ions on the growth and bioaccumulation properties of adapted and nonadapted biomass is investigated as a function of initial metal concentration. Binding capacity experiments using nonviable biomass are also performed as a function of temperature

RESULTS: The addition of Cu(II) and Cr(III) had a significant negative effect on the growth of yeast. Nonadapted cells could tolerate Cu(II) and Cr(III) ions up to a concentration of 75 ppm. The growth rate of nonadapted and adapted cells decreased with the increase in Cu(II) and Cr(III) concentration. Adapted *P. stipitis* biomass was capable of removing Cu(II) and Cr(III) with a maximum specific uptake capacity of 15.85 and 9.10 mg g⁻¹, respectively, at 100 ppm initial Cu(II) and Cr(III) concentration at pH 4.5. Adsorption data on nonviable cells were found to be well modeled by the Langmuir and Temkin isotherms. The maximum loading capacity of dry biomass predicted from Langmuir isotherm for Cu(II) and Cr(III) at 20 °C were 16.89 and 19.2 mg g⁻¹, respectively, at pH 4.5. Biosorptive capacities were dependent on temperature for Cu(II) and Cr(III) solutions.

CONCLUSION: Cu(II)- and Cr(III)-adapted cells grow and accumulate these ions at high ratios. On the other hand, nonviable *P. stipitis* was found to be an effective biosorbent for Cu(II) and Cr(III) biosorption.

Keywords: *Pichia stipitis* • bioaccumulation • biosorption • Cu(II) • Cr(III)

Geoffrey Michael Gadd *. (Division of Molecular and Environmental Microbiology, College of Life Sciences, University of Dundee, Dundee, DD1 5EH, Scotland, UK. email: [Geoffrey Michael Gadd \(g.m.gadd@dundee.ac.uk\)](mailto:Geoffrey.Michael.Gadd@dundee.ac.uk)) **Biosorption: critical review of scientific rationale, environmental importance and significance for pollution treatment. Journal of Chemical Technology & Biotechnology, Volume 84(1) (2009):13 - 28**

Biosorption may be simply defined as *the removal of substances from solution by biological material*. Such substances can be organic and inorganic, and in gaseous, soluble or insoluble forms. Biosorption is a physico-chemical process and includes such mechanisms as absorption, adsorption, ion exchange, surface complexation and precipitation. Biosorption is a property of both living and dead organisms (and their components) and has been heralded as a promising biotechnology for pollutant removal from solution, and/or pollutant recovery, for a number of years, because of its efficiency, simplicity, analogous operation to conventional ion exchange technology, and availability of biomass. Most biosorption studies have carried out on microbial systems, chiefly bacteria, microalgae and fungi, and with toxic metals and radionuclides, including actinides like uranium and thorium. However, practically all biological material has an affinity for metal species and a considerable amount of other research exists with macroalgae (seaweeds) as well as plant and animal biomass, waste organic sludges, and many other wastes or derived bio-products. While most biosorption research concerns metals and related substances, including radionuclides, the term is now applied to particulates and all manner of organic substances as well. However, despite continuing dramatic increases in published research on biosorption, there has been little or no exploitation in an industrial context. This article critically reviews aspects of biosorption research regarding the benefits, disadvantages, and future potential of biosorption as an industrial process, the rationale, scope and scientific value of biosorption research, and the significance of biosorption in other waste treatment processes and in the environment.

Keywords: biosorption • bioremediation • pollutants • toxic metals • radionuclides • organic wastes • dyes • bacteria • fungi • algae • biosorbent • adsorption

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BACKGROUND: Biosurfactant production was investigated using two strains of *Bacillus subtilis*, one being a reference strain (*B. subtilis* 1012) and the other a recombinant of this (*B. subtilis* W1012) made able to produce the green fluorescent protein (GFP).

RESULTS: Batch cultivations carried out at different initial levels of glucose (G_0) in the presence of 10 g L^{-1} casein demonstrated that the reference strain was able to release higher levels of biosurfactants in the medium at $5.0 \leq G_0 \leq 10 \text{ g L}^{-1}$ ($B_{\max} = 104\text{-}110 \text{ mg L}^{-1}$). The recombinant strain exhibited slightly lower levels of biosurfactants ($B_{\max} = 90\text{-}104 \text{ mg L}^{-1}$) but only at higher glucose concentrations ($G_0 \geq 20 \text{ g L}^{-1}$). Under these nutritional conditions, the

fluorescence intensity linked to the production of GFP was shown to be associated with the cell concentration even after achievement of the stationary phase.

CONCLUSION: The ability of the genetically-modified strain to simultaneously overproduce biosurfactant and GFP even at low biomass concentration makes it an interesting candidate for use as a biological indicator to monitor indirectly the biosurfactant production in bioremediation treatments.

Keywords: *Bacillus subtilis* • biomonitoring • biosurfactant production • GFP • casein • glucose

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Anatomical, histochemical and biochemical approaches were used to study mercury uptake and phytotoxicity as well as anti-oxidative responses in two species of ferns [Chinese brake fern (*Pteris vittata*) and Boston fern (*Nephrolepis exaltata*)], grown in a hydroponic system. The roots of both cultivars accumulated large amounts of mercury, but exhibited limited mercury translocation to shoots. Mercury exposure led to more pronounced phytotoxicity accompanied by stronger oxidative stress in the shoots of *P. vittata* than in *N. exaltata*. *N. exaltata* established a more effective anti-oxidative system against mercury-induced oxidative stress than did *P. vittata*. The activity of anti-oxidative enzymes (superoxide dismutase, catalase and glutathione reductase) increased. The reduced ascorbate (ASA) and oxidized ascorbate (DHA) are regulated. Mercury exposure led to an increase in the concentration of glutathione (GSH) in both fern species. The present study suggests that *N. exaltata* is more tolerant to mercury exposure than *P. vittata*, which has been also reported to be more tolerant to arsenic exposure. *N. exaltata* may thus have potential for phytostabilization of soils or phytofiltration of waste water contaminated with mercury.

Keywords: Mercury - Phytotoxicity - Oxidative stress - *Pteris vittata* - *Nephrolepis exaltata* - Phytoremediation

Bioremediation

Gulay Bayramoglu^a, Ihsan Gursel^b, Yagmur Tunali^c and M. Yakup Arica^a. (^aGazi University, Faculty of Arts and Sciences, Biochemical Processing and Biomaterial Research Laboratory, 06500 Teknik Okullar, Ankara, Turkey, ^bBilkent University, Molecular Biology and Genetics Department, Bilkent 06800, Ankara, Turkey, ^cAnadolu University, Faculty of Pharmacy, Department of Pharmaceutical Microbiology, 26470

Tepebaşı, Eskişehir, Turkey). Biosorption of phenol and 2-chlorophenol by *Funalia trogii* pellets. *Bioresource Technology*, Volume 100(10) (2009): 2685-2691

The removal of phenol (Ph) and 2-chlorophenol (2-CPh) from aqueous solution by native and heat inactivated fungus *Funalia trogii* pellets were investigated. The effects of contact time, solid/liquid ratio, optimum pH and temperature on the phenols removal capacity by the pellets were established. The removal efficiency of phenols increased significantly with increasing biomass dose. The optimum pH was detected to be 8.0. The second-order equations are described and evaluated on the basis of a comparative estimation of the corresponding coefficients. The phenol removal equilibrium isotherm was modeled by the Langmuir equations. The enthalpy change values were obtained between -7.62 and -10.64 kJ/mol. This indicated that the uptake of phenols either on native or heat inactivated fungal pellets was based on a physical adsorption process.

Keywords: *Funalia trogii*; Phenol; Bioremediation; Adsorption kinetic; Adsorption isotherm

Yi Cheng^a, Zhaohui Guo^a, Xueduan Liu^b, Huaqun Yin^b, Guanzhou Qiu^b, Fengkai Pan^a and Hongwei Liu^b. (^aInstitute of Environmental Engineering, School of Metallurgical Science and Engineering, Central South University, Changsha 410083, PR China, ^bDepartment of Bioengineering, School of Resources Processing and Bioengineering, Central South University, Changsha 410083, PR China). The bioleaching feasibility for Pb/Zn smelting slag and community characteristics of indigenous moderate-thermophilic bacteria. *Bioresource Technology*, Volume 100 (10) (2009): 2737-2740

The feasibility of recovering metal values and removing hazardous elements from the Pb/Zn smelting slag using bioleaching technique were studied through a flask experiment, and the community characteristics of the indigenous moderate-thermophilic bacteria in this bioleaching system were also analyzed through a culture-independent restriction fragment length polymorphism (RFLP) of 16S rRNA genes approach. The results show that more than 80% of Al, As, Cu, Mn, Fe and Zn in the Pb/Zn smelting slag were leached at 65 °C, pH 1.5, pulp density 5%, but only about 5% of Pb. Phylogenetic analysis revealed that the bacteria in the bioleaching system mainly fell among *Firmicutes*, *Gammaproteobacteria* and *Betaproteobacteria*, and the dominant bacteria are affiliated with *Bacillus* spp., *Sporosarcina* spp. and *Pseudomonas* spp.

Keywords: Moderate-thermophilic bacteria; Bioleaching; Pb/Zn smelting slag; 16S rRNA-RFLP; Phylogenetic analysis

Dalei Zhang^a, Hainan Kong^a, Deyi Wu^a, Shengbing He^a, Zhanbo Hu^{a, b} and Xiaofang Hu^a. (^aSchool of Environmental Science and Engineering, Shanghai JiaoTong University, Shanghai 200240, China, ^bSchool of Environment Studies, Guangxi University, Nanning 530004, China). Remediation of chromite ore processing residue by pyrolysis process with sewage sludge. *Bioresource Technology*, Volume 100(11) (2009): 2874-2877

The present work developed a novel technique to treat chromite ore processing residue (COPR). The process involved mixing the COPR with sewage sludge followed by pyrolysis. The gaseous organic fraction generated during pyrolysis of sludge was beneficial to Cr(VI) reduction. Process variables, such as the amount of sludge added to COPR (sludge-to-COPR (S/C) ratio), heating temperature, reaction time and particle size, were systematically varied, and their influences on

the Cr(VI) reduction in COPR were investigated. Cr(VI) content had decreased greatly, from 3384 mg kg⁻¹ for untreated COPR to less than 30 mg kg⁻¹ for COPR treated at 600 °C.

Keywords: Pyrolysis; COPR; Sewage sludge; Reduction

Slavomír Čerňanský^a, Marek Kolenčík^b, Jaroslav Ševc^b, Martin Urik^a and Edgar Hiller^c. (^aDepartment of Ecosozology and Physiotactics, Faculty of Natural Sciences, Comenius University in Bratislava, Mlynská dolina, 842 15 Bratislava, Slovakia, ^bInstitute of Geology, Faculty of Natural Sciences, Comenius University in Bratislava, Mlynská dolina, 842 15 Bratislava, Slovakia, ^cDepartment of Geochemistry, Faculty of Natural Sciences, Comenius University in Bratislava, Mlynská dolina, 842 15 Bratislava, Slovakia). **Fungal volatilization of trivalent and pentavalent arsenic under laboratory conditions. Bioresource Technology, Volume 100(2) (2009): 1037-1040**

Production of volatile derivatives of arsenic was studied using pure cultures of different fungal strains under laboratory conditions. Arsenic was used in its trivalent and pentavalent forms to evaluate the effect of arsenic valency on its biovolatilization. The average amount of volatilized arsenic for all fungal strains ranged from 0.026 mg to 0.257 mg and 0.024 mg to 0.191 mg of trivalent and pentavalent arsenic, respectively. These results show that approximately 23% of arsenic was volatilized from all culture media originally enriched with approximately 4 and 17 mg L⁻¹ of arsenic in trivalent form. The average amount of biovolatilized arsenic from culture media originally enriched with 4 and 17 mg L⁻¹ of arsenic in pentavalent form was 24% and 16%, respectively. The order of ability of arsenic biovolatilization is *Neosartorya fischeri* > *Aspergillus clavatus* > *Aspergillus niger*. Toxicity and fungal resistance to trivalent and pentavalent arsenic were also evaluated based on radial growth and biomass weight.

Keywords: Biovolatilization; Arsenic; Fungi; Toxicity

Rosa María Pérez Silva^a, Arelis Ábalos Rodríguez^a, José Manuel Gómez Montes De Oca^b and Domingo Cantero Moreno^b. (^aStudy Centre of Industrial Biotechnology, Faculty of Natural Sciences, University of East Santiago of Cuba, CP 90500, Cuba, ^bDepartment of Chemical Engineering, Food Technology and Environmental Technologies, University of Cádiz, CP11510 Puerto Real, Cádiz, Spain). **Biosorption of chromium, copper, manganese and zinc by *Pseudomonas aeruginosa* AT18 isolated from a site contaminated with petroleum. Bioresource Technology, Volume 100(4) (2009): 1533-1538**

The study describes the sorption of Cr, Cu, Mn and Zn by *Pseudomonas aeruginosa* AT18 isolated from a site contaminated with petroleum and heavy metals. The concentrations studied were 50, 49, 60 and 70 (mg L⁻¹) for Cr, Cu, Mn and Zn, respectively. The solution pH and ionic strength were very important factors in the metal biosorption performance and the biosorption capacity of *P. aeruginosa* AT18 for Cr³⁺, Cu²⁺, Mn²⁺ and Zn²⁺. In aqueous solution, the biosorption increased with increasing pH in the range 5.46–7.72. The results obtained in the experimental assays show that *P. aeruginosa* AT18 has the capacity for biosorption of the metallic ions Cr³⁺, Cu²⁺ and Zn²⁺ in solutions, although its capacity for the sorption of manganese is low (22.39 mg Mn²⁺/g of biomass) in comparison to the Cr³⁺, Cu²⁺ and Zn²⁺ ions, as shown by the individual analyses. However, 20% of the manganese was removed from an initial concentration of 49.0 mg L⁻¹, with a Q_m value similar to that obtained in solutions

containing mixtures of Cr^{3+} , Cu^{2+} , Mn^{2+} and Zn^{2+} . The chromium level sorbed by *P. aeruginosa* AT18 biomass was higher than that for Cu, Mn and Zn, with 100% removal in the pH range 7.00–7.72 and a Q_m of 121.90–200.00 mg of Cr^{3+} /g of biomass. The removal of Cr, Cu and Zn is also a result of precipitation processes.

Keywords: *Pseudomonas aeruginosa*; Heavy metals; Biosorption

Han Chen^a, Yu-Bei Cai^a, Wen-Juan Zhang^a and Wei Li^a. (^aDepartment of Environmental Engineering, Zhejiang University (Yuquan Campus), Hangzhou 310027, China). **Methoxylation pathway in biodesulfurization of model organosulfur compounds with *Mycobacterium* sp. *Bioresource Technology*, Volume 100(6) (2009): 2085-2087**

A metabolic pathway for the biodesulfurization of model organosulfur compounds e.g., dibenzothiophene (DBT), is proposed. This pathway, defined as extended 4S pathway, incorporates the traditional 4S pathway with the methoxylation pathway from 2-hydroxybiphenyl (HBP) to 2-methoxybiphenyl (2-MBP). The formation of 2-MBP was confirmed by the gas chromatography–mass spectrometry (GC–MS) analysis. A similar pathway was also obtained in the desulfurization of 4,6-dimethyldibenzothiophene (4,6-DMDBT), confirming the methoxylation reaction in the desulfurization process by the *Mycobacterium* sp. strain. Compared with 2-HBP, 2-MBP has much slighter inhibition effect on the cell growth and desulfurization activity. Thus, the methoxylation pathway from 2-HBP to 2-MBP would make less inhibitory effect on the microbe. The new pathway with 2-MBP as the end product may be an alternative for the further desulfuration of the fossil fuels.

Keywords: Biodesulfurization; Pathway; Dibenzothiophene; Methoxylation; *Mycobacterium* sp.

Atac Uzel^a and Guven Ozdemir^a. (^aEge University, Faculty of Sciences, Department of Biology, Basic and Industrial Microbiology Section, 35100 Bornova-Izmir, Turkey). **Metal biosorption capacity of the organic solvent tolerant *Pseudomonas fluorescens* TEM08. *Bioresource Technology*, Volume 100(2) (2009): 542-548**

Many kinds of biomass are being tested as a biosorption material for metal removal from the contaminated waters. In the present study the biosorption capacity of an organic solvent tolerant (OST) bacterium was investigated against Cr(VI) and Ni(II). The OST strain of *Pseudomonas fluorescens* TEM08 was isolated from an oil contaminated soil sample and grown in normal culture conditions (type I) and in the presence of the cyclohexane (type II). Two types of cells were used in the biosorption experiments to compare the organic solvent effect on the biosorption capacity. The biosorption equilibrium was described by Langmuir and Freundlich adsorption isotherms. The value of Q^0 was higher for type I cells (40.8 for Cr(VI); 12.4 for Ni(II)) than the type II (40.7 for Cr(VI); 11.2 for Ni(II)). The adsorption capacity constants (K_F) of Freundlich model for type I cells and for type II cells were 10.87 and 8.78 for Ni(II) and 13.60 and 10.99 for Cr(VI), respectively.

Keywords: Biosorption; Organic solvent tolerant; *Pseudomonas fluorescens*; Chromium; Nickel

Thyagarajan Mathialagan^a and Thiruvencatachari Viraraghavan^b. (^aCity of Abbotsford, Surrey, BC, Canada V3W 8A4, ^bFaculty of Engineering, University of Regina, Regina, SK, Canada S4S 0A2). **Biosorption of pentachlorophenol from aqueous solutions by a fungal biomass. *Bioresource Technology*, Volume 100(2) (2009): 549-558**

This study focuses on the use of non-viable *Aspergillus niger* biomass, for the biosorption of pentachlorophenol (PCP) from aqueous solutions. Various forms of the biomass-autoclaved and chemically conditioned, were tested for their potential in the removal of PCP from aqueous solutions. It was found that PCP removal was pH dependent; PCP removal decreased with the increase in pH for all type of biomass, except for cetyltrimethylammonium bromide (CTAB) biomass. For CTAB biomass, a near complete removal of PCP was observed at all pHs. Therefore, CTAB biomass was used in further studies. PCP removal was rapid, with an equilibrium time of 2 h. The rate of adsorption kinetics was well described by a pseudo-second order model. Isotherm models of the type one and two parameter models were found to fit the isotherm data. PCP biosorption was found to be exothermic in nature; the amount of PCP sorbed decreased with an increase in temperature. Desorption was carried out using deionized water, dilute HCl and dilute NaOH, and it was found that most of the PCP was irreversibly bound to the biomass. The addition of inorganic salts did not affect the removal of PCP from aqueous solutions. Among the surface functional groups present on the biomass, carboxyl, amide and hydroxyl groups seem to have played a role in PCP biosorption. It was concluded that CTAB treated biomass was an excellent adsorbent for the removal of PCP from aqueous solutions.

Keywords: Pentachlorophenol; *Aspergillus niger*; Kinetics; Isotherms; Desorption

Hisashi Saeki^a, Masaru Sasaki^a, Koei Komatsu^a, Akira Miura^b and Hitoshi Matsuda^a. (^aBio Research Center, Japan Energy Corporation, 3-17-35 Niizo-minami, Toda-shi, Saitama 335-0027, Japan, ^bTechnology Development Center, Nippon Mining & Metals Co. Ltd., 1-1-2 Shirogane-cho, Hitachi-shi, Ibaraki 317-0056, Japan). **Oil spill remediation by using the remediation agent JE1058BS that contains a biosurfactant produced by *Gordonia* sp. strain JE-1058. *Bioresource Technology*, Volume 100(2) (2009): 572-577**

A remediation agent containing a biosurfactant was prepared by spray drying the sterilized culture broth of *Gordonia* sp. strain JE-1058, and the agent was designated as JE1058BS. On subjection to the baffled flask test developed by the United States Environmental Protection Agency, JE1058BS showed a strong potential to be applied as an oil spill dispersant even in the absence of a solvent. It also proved to be an effective bioremediation agent for the remediation of oil spills at sea. The addition of JE1058BS to seawater stimulated the degradation of weathered crude oil (ANS 521) via the activity of the indigenous marine bacteria. Its addition also stimulated the removal of crude oil from the surface of contaminated sea sand. These results indicate that biosurfactant-containing JE1058BS has a strong potential to be applied as a remediation agent for the clean-up of oil spills at sea and on shorelines.

Keywords: Dispersants; Biosurfactant; Bioremediation agent; JE1058BS; *Gordonia*

Hong-Bo Zhou^{a, b}, Wei-Min Zeng^{a, b}, Zhi-Feng Yang^a, Ying-Jian Xie^a and Guan-Zhou Qiu^{a, b}. (^aSchool of Minerals Processing and Bioengineering, Central South University, Changsha 410083, China, ^bKey Laboratory of Biometallurgy, Ministry of Education, Changsha, Hunan 410083, China). **Bioleaching of chalcopyrite concentrate by a moderately thermophilic culture in a stirred tank reactor. *Bioresource Technology*, Volume 100(2) (2009): 515-520**

A mixed culture of moderately thermophilic microorganisms was enriched from acid mine drainage samples collected from several chalcopyrite mines in China. Such mixed culture can be used to effectively extract copper from chalcopyrite. Furthermore, after being adapted to gradually increased concentration of chalcopyrite concentrate, the tolerance of the mixed culture to chalcopyrite concentrate was brought up to 80 g/L. The effects of several leaching parameters on copper recovery in stirred tank reactor also had been investigated. The results of the investigation show that it was possible to achieve a copper extraction rate of 75% in 44 days at a pulp density of 8%. The leaching rate of chalcopyrite concentrate tended to increase with dissolved total iron concentration. At low pH ranges, more microscopic counts of microorganisms were found in the solution. Furthermore, the analysis of leached residues indicates that the passivation of chalcopyrite concentrate was mainly due to a mass of jarosite and PbSO_4 on the mineral surface, other than the elemental sulphur layer. The bacterial community composition was analyzed by using Amplified Ribosomal DNA Restriction Analysis. Two moderately thermophilic bacteria species were identified as *Leptospirillum ferriphilum* and *Acidithiobacillus caldus* with abundance of 67% and 33% in the bio-pulp, respectively.

Keywords: Moderately thermophilic microorganisms; Stirred tank reactor; Chalcopyrite concentrate; Community composition

Piyush Kant Pandey^a, Shweta Choubey^a, Yashu Verma^a, Madhurima Pandey^a and K. Chandrashekar^b. (^aCentre for Environmental Science and Engineering, Department of Engineering Chemistry, Bhilai Institute of Technology, Durg 491002, CG, India, ^bAnalytical Chemistry Group, Defence Metallurgical Research Laboratory (DMRL), Hyderabad 500058, AP, India). **Biosorptive removal of arsenic from drinking water. Bioresource Technology, Volume 100(2) (2009): 634-637**

A biomass derived from the plant *Momordica charantia* has been found to be very efficient in arsenic(III) adsorption. An attempt was made to use this biomass for arsenic(III) removal under different conditions. The parameters optimized were contact time (5–150 min), pH (2–11), concentration of adsorbent (1–50 g/l), concentration of adsorbate (0.1–100 mg/l), etc. It was observed that the pH had a strong effect on biosorption capacity. The optimum pH obtained for arsenic adsorption was 9. The influence of common ions such as Ca^{2+} , Mg^{2+} , Cd^{2+} , Se^{4+} , Cl^- , SO_4^{2-} , and HCO_3^- , at concentrations varying from 5 to 1000 mg/l was investigated. To establish the most appropriate correlation for the equilibrium curves, isotherm studies were performed for As(III) ion using Freundlich and Langmuir adsorption isotherms. The pattern of adsorption fitted well with both models. The biomass of *M. charantia* was found to be effective for the removal of As(III) with 88% sorption efficiency at a concentration of 0.5 mg/l of As(III) solution, and thus uptake capacity is 0.88 mg As(III)/gm of biomass. It appears that this biomass should be used as a palliative food item. Further it also appears that the dietary habits may play a role in the toxic effects of ingested arsenic.

Keywords: Arsenic; Adsorption; Biomass; *Momordica charantia*

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^dDepartment of Biochemistry, Faculty of Science, Tarbiat Modares University, Tehran, Iran). Biodesulfurization of dibenzothiophene by recombinant *Gordonia alkanivorans* RIPI90A. *Bioresource Technology*, Volume 100(1) (2009): 475-479

The *dszABC* genes from newly reported dibenzothiophene biodesulfurizing bacterium, *Gordonia alkanivorans* RIPI90A were cloned and sequenced. The overall nucleotide sequence similarity between the *dszABC* genes of *G. alkanivorans* RIPI90A and those of *Rhodococcus erythropolis* IGTS8 and *Gordonia nitida* were 83.1% and 83.2%, respectively. A gene transfer system for *G. alkanivorans* RIPI90A was established employing the *Escherichia coli*-*Rhodococcus* shuttle vector pRSG43 as suitable cloning vector, resulting in transformation efficiencies up to 1.6×10^5 CFUs μg^{-1} plasmid DNA. This stable vector was applied to cloning and efficient expression of the *dsz* genes under the control of *lac* promoter. The recombinant strain was able to desulfurize dibenzothiophene in the presence of inorganic sulfate and sulfur-containing amino acids. The maximum desulfurization activity by recombinant resting cells ($131.8 \mu\text{M}$ 2-hydroxybiphenyl $\text{g}^{-1}_{\text{dry cell weight}} \text{h}^{-1}$) was increased 2.67-fold in comparison to the highest desulfurization activity of native resting cells.

Keywords: Biodesulfurization; *Gordonia*; *dsz* genes; Recombinant bacteria

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The bacterial leaching of zinc and iron from solid wastes at the Isdemir iron and steel plant has been investigated using *Acidithiobacillus ferrooxidans* as the bacterial agent. The effects of a range of operational parameters, including particle size, solids concentration and pH, on the efficiency of the bioleaching process were investigated. In each test, several variables were determined to assess the efficiency of leaching, including slurry pH and redox potential, temperature, bacteria population and concentrations of zinc and iron in solution. Experimental results demonstrated that pulp solids concentration, slurry pH and solids particle size were all important parameters in the bacterial leaching process. Maximum extraction was achieved at pH values around 1.3 and a solids concentration of 1% w/v, with 35% of the Zn content and 37% of the Fe being dissolved.

Keywords: Iron ores - Tailings - Bacteria - Bioleaching - Recycling - Waste processing - *Acidithiobacillus ferrooxidans*

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in Residual Soybean Oil. Applied Biochemistry and Biotechnology, Volume 152(1) (2009): 156-168

Pseudomonas aeruginosa PACL strain, isolated from oil-contaminated soil taken from a lagoon, was used to investigate the efficiency and magnitude of biosurfactant production, using different waste frying soybean oils, by submerged fermentation in stirred tank reactors of 6 and 10 l capacities. A complete factorial experimental design was used, with the goal of optimizing the aeration rate (0.5, 1.0, and 1.5 vvm) and agitation speed (300, 550, and 800 rpm). Aeration was identified as the primary variable affecting the process, with a maximum rhamnolipid concentration occurring at an aeration rate of 0.5 vvm. At optimum levels, a maximum rhamnolipid concentration of 3.3 g/l, an emulsification index of 100%, and a minimum surface tension of 26.0 dynes/cm were achieved. Under these conditions, the biosurfactant production derived from using a mixture of waste frying soybean oil (WFSO) as a carbon source was compared to production when non-used soybean oil (NUSO), or waste soybean oils used to fry specific foods, were used. NUSO produced the highest level of rhamnolipids, although the waste soybean oils also resulted in biosurfactant production of 75–90% of the maximum value. Under ideal conditions, the kinetic behavior and the modeling of the rhamnolipid production, nutrient consumption, and cellular growth were established. The resulting model predicted data points that corresponded well to the empirical information.

Keywords: Biosurfactants - Glycolipids - *Pseudomonas aeruginosa* - Rhamnolipids - Surface-active compounds - Soybean oil

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The production of cellulolytic enzymes by the fungus *Aspergillus phoenicis* was investigated. Grape waste from the winemaking industry was chosen as the growth substrate among several agro-industrial byproducts. A 2²—2 central composite design was performed, utilizing the amount of grape waste and peptone as independent variables. The fungus was cultivated in submerged fermentation at 30  C and 120 rpm for 120 h, and the activities of total cellulases, endoglucanases, and  -glucosidases were measured. Total cellulases were positively influenced by the linear increase of peptone concentration and decrease at axial concentrations of grape waste and peptone. Maximum activity of endoglucanase was observed by a linear increase of both grape waste and peptone concentrations. Concentrations of grape waste between 5 and 15 g/L had a positive effect on the production of  -glucosidase; peptone had no significant effects. The optimum production of the three cellulolytic activities was observed at values near the central point. *A. phoenicis* has the potential for the production of cellulases utilizing grape waste as the growth substrate.

Keywords Agro-industrial waste - Factorial design - Cellulase -  -Glucosidases - *Aspergillus phoenicis*

Aike C. da Silva¹, Fernando J. S. de Oliveira², Diogo S. Bernardes¹ and Francisca P. de Fransa¹. (¹Escola de Qumica, Universidade Federal do Rio de Janeiro, Centro de Tecnologia, Bloco E, Ilha do Fundo, 21949-900 Rio de Janeiro, RJ, Brazil, ²Petrleo Brasileiro SA, Avenida Almirante Barroso, 81, 24 andar, Centro, 20031-004 Rio de Janeiro, RJ, Brazil). **Bioremediation of Marine Sediments Impacted by Petroleum. Applied Biochemistry and Biotechnology, Volume 153(1-3) (2009): 58-66**

The aim of this work was to optimize the bioremediation of crude oil-contaminated sand sediment through the biostimulation technique. The soil was obtained in the mid-tide zone of Guanabara Bay, Rio de Janeiro, Brazil and was artificially contaminated with crude oil at 14 g kg⁻¹. Bioremediation optimization was performed using an experimental design and statistical analysis of the following factors: supplementation with commercial biosurfactant Jeneil IBR 425 and commercial mineral NPK fertilizer. The response variable used was the biodegradation of the heavy oil fraction, HOF. The analysis of the studied factors and their interactions was executed using contour plots, Pareto diagram and ANOVA table. Experimental design results indicated that the supplementation with fertilizer at 100:25:25 C/N/P ratio and biosurfactant at 2 g kg⁻¹ yielded biodegradation of HOF at about 30% during 30 days of process. Some experiments were carried out using the experimental design results, yielding 65% of biodegradation of HOF and 100% of *n*- alkanes between C15 and C30 during 60 process days. Intrinsic biodegradation test was carried out, yielding 85% of biodegradation of *n*-alkanes between C15 and C30 during 30 days of process.

Keywords: Marine sediments - Petroleum - Bioremediation - Biostimulation - Intrinsic bioremediation

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The bioremediation of the nematicide oxamyl, applied at the recommended rate of 6 l ha⁻¹ in sandy soil cultivated with tomato and amended with different animal manures at the recommended dose of 2.5 tons ha⁻¹, was investigated. The experiment was conducted in a controlled environmental chamber under a 16-h photoperiod, with a light intensity of 300 μ Em⁻² s⁻¹ at 25 °C and relative humidity of 70 \pm 5%. The remaining amount of oxamyl in soil was extracted after different time intervals based on the solid phase extraction (SPE) with methanol and then analyzed by HPLC. Only the peak corresponding to oxamyl was observed in the chromatogram and no intermediate could be detected. By the end of the experiment (28 days), the dissipation percentage of oxamyl reached about 99% in the case of bovine manure-amended soil. This rate of disappearance was 1.76 times higher than in unamended-soil, while poultry and sheep manures enhanced the dissipation rate by 1.52 and 1.44 times, respectively. The disappearance rate constants and half-life values of the compound were obtained from the exponential decay equations. The decomposition of oxamyl in the control followed the first order kinetics with $t_{1/2}$ of about 26 days. On the other hand, a biphasic model was assumed to

explore the disappearance of oxamyl in soil amended with different animal manures where the rate of disappearance in the first phase was faster than the second phase. This is clearly reflected in the half-life ($t_{1/2}$) values for the first and second phases, where the $t_{1/2}$ values of oxamyl ranged from 3.19 to 5.41 and 9.76 to 43.31 days, respectively. The results demonstrated that animal manures may offer an efficient, cheap, safe, and friendly bioremediator for pesticide-polluted soil.

Keywords: Oxamyl; Animal manures; Bioremediation; Solid phase extraction; Half-life

Marek Koutny^a, Pierre Amato^b, Marketa Muchova^a, Jan Ruzicka^a and Anne-Marie Delort^b. (^aDepartment of Environmental Protection Engineering, Faculty of Technology, Tomas Bata University in Zlin, TGM sq. 275, 762 72 Zlin, Czech Republic, ^bLaboratoire de Synthèse Et Etude de Systemes a Interet Biologique (SEESIB), UMR 6504 CNRS, Ensemble Universitaire des Cezeaux, Universite Blaise Pascal, 63 177 Aubiere Cedex, France). **Soil bacterial strains able to grow on the surface of oxidized polyethylene film containing prooxidant additives. International Biodeterioration & Biodegradation, Volume 63(3) (2009): 354-357**

Twelve bacterial strains able to adsorb and grow on the surface of oxidized low-density polyethylene film containing prooxidant additives were isolated from three forest soils and subsequently identified. Most of them belonged to different genera of the proteobacteria group; however, three of the isolates were *Rhodococcus* strains. With the exception of one of the *Rhodococcus* strains, the isolates did not exhibit significant hydrophobicity of their cell surfaces. The study showed that bacteria capable of adhering to the surface of oxidized polyethylene, growing there and possibly biodegrading its oxidation products are not rare in forest soils and that they belong to different taxonomical groups common in soil environment.

Keywords: Polyethylene; Biodegradation; Soil; Prooxidants; Bacteria; Hydrophobicity

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New amphiphilic block surfactants ABA based on a central segment of polycaprolactone with different molecular composition were evaluated in the bioremediation of naphthalene in water by *Sphingomonas paucimobilis* and compared with sodium dodecyl sulphate as reference surfactant (SDS). Also the biodegradation of the new surfactants by bacteria, *S. paucimobilis* and a mixture of bacteria (*Pseudomonas aureginosa*, *Bacillus subtilis*, *Bacillus amyloliquefaciens* and *Bacillus megaterium*) was studied by indirect impedance technique and carbon dioxide determination. All the bacteria biodegraded in solution and micellar phase the central segment of PCL with mineralization rates in the range of 0.024–0.036 mg of CO₂ per day.

S. paucimobilis biodegraded naphthalene in the presence of the new surfactants and GC analysis demonstrated that conversion to products started immediately after inoculum. In all the experiments, except for SDS, at 140 h of incubation time, the remaining naphthalene concentration was about 10% of the initial concentration. In contrast, the production of CO₂ was delayed 4–7 days and values around 75% of naphthalene mineralization degree were achieved in

three weeks. The addition of PCL-surfactants, in solution and in micellar phase, not interfered in the naphthalene mineralization. These results have shown promising potential of these biodegradable PCL-surfactants in surfactant-enhanced remediation (SER) technology for removing residual organics from contaminated groundwater and soils.

Keywords: Biodegradation; Bioremediation; Surfactants; Polycaprolactone; *Pseudomonas*; *Bacillus*; Indirect impedance technique; Naphthalene

Jianlong Wang^a and Can Chen^a. (^aLaboratory of Environmental Technology, INET, Tsinghua University, Beijing 100084, PR China). Biosorbents for heavy metals removal and their future. *Biotechnology Advances*, Volume 27(2) (2009): 95-226

A vast array of biological materials, especially bacteria, algae, yeasts and fungi have received increasing attention for heavy metal removal and recovery due to their good performance, low cost and large available quantities. The biosorbent, unlike mono functional ion exchange resins, contains variety of functional sites including carboxyl, imidazole, sulphhydryl, amino, phosphate, sulfate, thioether, phenol, carbonyl, amide and hydroxyl moieties. Biosorbents are cheaper, more effective alternatives for the removal of metallic elements, especially heavy metals from aqueous solution. In this paper, based on the literatures and our research results, the biosorbents widely used for heavy metal removal were reviewed, mainly focusing on their cellular structure, biosorption performance, their pretreatment, modification, regeneration/reuse, modeling of biosorption (isotherm and kinetic models), the development of novel biosorbents, their evaluation, potential application and future. The pretreatment and modification of biosorbents aiming to improve their sorption capacity was introduced and evaluated. Molecular biotechnology is a potent tool to elucidate the mechanisms at molecular level, and to construct engineered organisms with higher biosorption capacity and selectivity for the objective metal ions. The potential application of biosorption and biosorbents was discussed. Although the biosorption application is facing the great challenge, there are two trends for the development of the biosorption process for metal removal. One trend is to use hybrid technology for pollutants removal, especially using living cells. Another trend is to develop the commercial biosorbents using immobilization technology, and to improve the biosorption process including regeneration/reuse, making the biosorbents just like a kind of ion exchange resin, as well as to exploit the market with great endeavor.

Keywords: Biosorbent; Biosorption; Heavy metal ions; Bacteria; Fungi; Algae; Biomass; Kinetics; Immobilization; Application

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There are strong drivers to increasingly adopt bioremediation as an effective technique for risk reduction of hydrocarbon impacted soils. Researchers often rely solely on chemical data to assess bioremediation efficiently, without making use of the numerous biological techniques for assessing microbial performance. Where used, laboratory experiments must be effectively

extrapolated to the field scale. The aim of this research was to test laboratory derived data and move to the field scale. In this research, the remediation of over thirty hydrocarbon sites was studied in the laboratory using a range of analytical techniques. At elevated concentrations, the rate of degradation was best described by respiration and the total hydrocarbon concentration in soil. The number of bacterial degraders and heterotrophs as well as quantification of the bioavailable fraction allowed an estimation of how bioremediation would progress. The response of microbial biosensors proved a useful predictor of bioremediation in the absence of other microbial data. Field-scale trials on average took three times as long to reach the same endpoint as the laboratory trial. It is essential that practitioners justify the nature and frequency of sampling when managing remediation projects and estimations can be made using laboratory derived data. The value of bioremediation will be realised when those that practice the technology can offer transparent lines of evidence to explain their decisions.

Detailed biological, chemical and physical characterisation reduces uncertainty in predicting bioremediation.

Keywords: Hydrocarbons; Bioremediation; Respiration; Laboratory-scale; Field-scale; Degradation; Microbial biosensors

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Pteris vittata plants were grown on twenty-one UK soils contaminated with arsenic (As) from a wide range of natural and anthropogenic sources. Arsenic concentration was measured in fern fronds, soil and soil pore water collected with *Rhizon* samplers. Isotopically exchangeable soil arsenate was determined by equilibration with ⁷³As^V. Removal of As from the 21 soils by three sequential crops of *P. vittata* ranged between 0.1 and 13% of total soil As. Ferns grown on a soil subjected to long-term sewage sludge application showed reduced uptake of As because of high available phosphate concentrations. A combined solubility-uptake model was parameterised to enable prediction of phytoremediation success from estimates of soil As, 'As-lability' and soil pH. The model was used to demonstrate the remediation potential of *P. vittata* under different soil conditions and with contrasting assumptions regarding re-supply of the labile As pool from unavailable forms.

This paper presents a predictive model for phytoremediation of soils, historically contaminated with arsenic, by the hyperaccumulator *P. vittata*.

Keywords: Arsenic; Chinese Brake fern; Phytoremediation; *Pteris vittata*

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Street, Liverpool, L3 3AF, United Kingdom). Phytoremediation trials on metal- and arsenic-contaminated pyrite wastes (Torviscosa, Italy). Environmental Pollution, Volume 157(3) (2009): 887-894

At a site in Udine, Italy, a 0.7 m layer of As, Co, Cu, Pb and Zn contaminated wastes derived from mineral roasting for sulphur extraction had been covered with an unpolluted 0.15 m layer of gravelly soil. This study investigates whether woody biomass phytoremediation is a realistic management option. Comparing ploughing and subsoiling (0.35 m depth), the growth of *Populus* and *Salix* and trace element uptake were investigated in both pot and field trials. Species differences were marginal and species selection was not critical. Impaired above-ground productivity and low translocation of trace elements showed that bioavailable contaminant stripping was not feasible. The most significant finding was of coarse and fine roots proliferation in surface layers that provided a significant sink for trace elements. We conclude that phytostabilisation and effective immobilisation of metals and As could be achieved at the site by soil amelioration combined with woody species establishment. Confidence to achieve a long-term and sustainable remediation requires a more complete quantification of root dynamics and a better understanding of rhizosphere processes.

In As- and metal-contaminated pyrite wastes, contaminant stripping is not feasible, and root foraging and quantification of root dynamics holds the key to stabilisation in woody species.

Keywords: Trace metals; Phytoremediation; Poplar; Pyrite cinders; Root electrical capacitance; Root growth; Willow

Abbreviations: CRW, coarse root weight; EC, root electrical capacitance; FRW, fine root weight; HM, heavy metals; RLD, volumetric root length density; SW, shoot weight; TRW, total root weight

Susan Tandy^a, John R. Healey^a, Mark A. Nason^a, Julie C. Williamson^a and Davey L. Jones^a. (^aSchool of the Environment and Natural Resources, Bangor University, Gwynedd LL57 2UW, UK). **Remediation of metal polluted mine soil with compost: Co-composting versus incorporation. Environmental Pollution, Volume 157(2) (2009): 690-697**

Trace element contamination of post-industrial sites represents a major environmental problem and sustainable management options for remediating them are required. This study compared two strategies for immobilizing trace elements (Cu, Pb, Zn, and As) in mine spoil: (1) co-composting contaminated soil with organic wastes and (2) conventional incorporation of mature compost into contaminated soil. Sequential chemical extraction of the soil was performed to determine temporal changes in trace element fractionation and bioavailability during composting and plant growth. We show that mine spoil can be co-composted successfully and this action causes significant shifts in metal availability. However, co-composting did not lead to significant differences in metal partitioning in soil or in plant metal uptake compared with simply mixing mine spoil with mature compost. Both treatments promoted plant growth and reduced metal accumulation in plants. We conclude that co-composting provides little additional benefit for remediating trace-element-polluted soil compared with incorporation of compost.

Co-composting did not provide enhanced stabilization of trace elements over the conventional addition of compost to contaminated land.

Keywords: Dissolved organic matter; Heavy metal stabilization; Mine rehabilitation; Phytoremediation; Site restoration; Soil pollution

Silke Nissen^a, Bruce D. Alexander^b, Ilyas Dawood^c, Martin Tillotson^c, Richard P.K. Wells^b, Donald E. Macphee^b and Kenneth Killham^a. (^aDepartment of Plant and Soil Science, Cruickshank Building, University of Aberdeen, St. Machar Drive, Aberdeen AB24 3UU, Scotland, UK, ^bDepartment of Chemistry, University of Aberdeen, Meston Walk, Aberdeen AB24 3UE, Scotland, UK, ^cYorkshire Water Services Ltd., Western House, Halifax Road, Bradford BD6 2SZ, UK). **Remediation of a chlorinated aromatic hydrocarbon in water by photoelectrocatalysis. Environmental Pollution, Volume 157(1) (2009): 72-76**

Photoelectrocatalysis driven by visible light offers a new and potentially powerful technology for the remediation of water contaminated by organo-xenobiotics. In this study, the performance of a visible light-driven photoelectrocatalytic (PEC) batch reactor, applying a tungsten trioxide (WO₃) photoelectrode, to degrade the model pollutant 2,4-dichlorophenol (2,4-DCP) was monitored both by toxicological assessment (biosensing) and chemical analysis. The bacterial biosensor used to assess the presence of toxicity of the parent molecule and its breakdown products was a multicopy plasmid *lux*-marked *E. coli* HB101 pUCD607. The bacterial biosensor traced the removal of 2,4-DCP, and in some case, its toxicity response suggests the identification of transient toxic intermediates. The loss of the parent molecule, 2,4-DCP determined by HPLC, corresponded to the recorded photocurrents. Photoelectrocatalysis offers considerable potential for the remediation of chlorinated hydrocarbons, and that the biosensor based toxicity results identified likely compatibility of this technology with conventional, biological wastewater treatment.

Visible light-driven photoelectrocatalysis has potential as a remediation technology in wastewater treatment.

Keywords: 2,4-Dichlorophenol; Photoelectrocatalysis; Bacterial biosensor; Wastewater; Tungsten trioxide; Photocatalyst

Doris Krpata^a, Walter Fitz^b, Ursula Peintner^a, Ingrid Langer^b and Peter Schweiger^b. (^aInstitute of Microbiology, Innsbruck University, Technikerstraße 25, A-6020 Innsbruck, Austria, ^bInstitute of Soil Science, University of Natural Resources and Applied Life Sciences, Peter Jordan-Straße 82, A-1190 Vienna, Austria). **Bioconcentration of zinc and cadmium in ectomycorrhizal fungi and associated aspen trees as affected by level of pollution. Environmental Pollution, Volume 157(1) (2009): 280-286**

Concentrations of Zn and Cd were measured in fruitbodies of ectomycorrhizal (ECM) fungi and leaves of co-occurring accumulator aspen. Samples were taken on three metal-polluted sites and one control site. Fungal bioconcentration factors (BCF = fruitbody concentration: soil concentration) were calculated on the basis of total metal concentrations in surface soil horizons (BCF_{tot}) and NH₄NO₃-extractable metal concentrations in mineral soil (BCF_{lab}). When plotted on log-log scale, values of BCF decreased linearly with increasing soil metal concentrations. BCF_{lab} for both Zn and Cd described the data more closely than BCF_{tot}. Fungal genera differed in

ZnBCF but not in CdBCF. The information on differences between fungi with respect to their predominant occurrence in different soil horizons did not improve relations of BCF with soil metal concentrations. Aspen trees accumulated Zn and Cd to similar concentrations as the ECM fungi. Apparently, the fungi did not act as an effective barrier against aspen metal uptake by retaining the metals.

Populus tremula and associated ectomycorrhizal fungi accumulate zinc and cadmium to similar concentrations.

Keywords: Metals; Accumulation; Ectomycorrhizas; Fruitbodies; *Populus tremula*

Ilse Forrez, Marta Carballa, Nico Boon, Willy Verstraete * (Laboratory of Microbial Ecology and Technology (LabMET), Faculty of Bioscience Engineering, Ghent University, Coupure Links 653, B-9000 Gent, Belgium. email: Willy Verstraete (Willy.Verstraete@UGent.be). **Biological removal of 17 α -ethinylestradiol (EE2) in an aerated nitrifying fixed bed reactor during ammonium starvation. Journal of Chemical Technology & Biotechnology, Volume 84(1) (2009):119 - 125**

BACKGROUND: Conventional wastewater treatment plants (WWTPs) tend to partially remove recalcitrant chemicals, such as pharmaceuticals. Among these, the synthetic estrogen 17 α -ethinylestradiol (EE2) is of great environmental concern. In this work a continuously aerated submerged fixed bed bioreactor was used for the biological removal of EE2 at $\mu\text{g L}^{-1}$ levels.

RESULTS: Removal efficiencies higher than 96% were obtained at a hydraulic retention time (HRT) of 4.3 days and a volumetric loading rate (B_v) of $11 \mu\text{g EE2 L}^{-1} \text{d}^{-1}$. Increasing the B_v up to 40 and $143 \mu\text{g EE2 L}^{-1} \text{d}^{-1}$ led to slightly lower removal efficiencies, 81 and 74%, respectively. Nitrification was confirmed to be the main biological mechanism involved in EE2 removal. Most interestingly, the elimination of EE2 was not affected by the absence of ammonium in the feed, suggesting that ammonia-oxidizing bacteria (AOB) were able to maintain their population density and their activity, even after several months of starvation.

CONCLUSION: The concept of an aerated submerged fixed bed bioreactor, capable of removing estrogens in a sustainable and biological way, shows great potential as an effluent polishing step for existing WWTPs.

Keywords: ammonium monooxygenase • biodegradation • estrogens • micropollutants • nitrification • post-treatment

T. Endreny^a and V. Collins^a. (^a423 Baker Labs, 1 Forestry Drive, SUNY ESF, Syracuse, NY 13210, United States). **Implications of bioretention basin spatial arrangements on stormwater recharge and groundwater mounding. Ecological Engineering, Volume 35(5) (2009): 670-677**

Stormwater bioretention basin recharge has the potential to raise the watertable and adversely impact subsurface infrastructure, undermining the benefits of naturalizing the urban water cycle. This research examined how groundwater mounding responded to three spatial arrangements of bioretention basins, from separated units to clustered units to single units, and changes in

hydraulic conductivity, storm intensity, and antecedent recharge, for 28 sub-watersheds in an 8-ha Syracuse, New York, sewershed with 43% impervious cover. Bioretention basin volumetric capacities were designed for a 24-h duration 2-yr return interval rainfall event. MODFLOW simulations with hydraulic conductivity at 1 cm h^{-1} predicted an increase in median groundwater mounding from 0.28 m to 0.72 m when separation distances were reduced from equally distributed to single units. In sag points, however mounding exceeded 1 m. By setting hydraulic conductivity to 0.01 cm h^{-1} , a worst case scenario, median mounding was greater than 1 m for all spatial designs, in all locations. Groundwater mound overlap was identified for spatial arrangements where intersecting streets created superposition, and greater mounding was observed at corner-situated bioretention basins. After 30 years of recharge, the steady state watertable had risen by 1.1 m, and subsequent storm event mounding could interfere with subsurface infrastructure in approximately 20% of the watershed, localized in the floodplain. This study recommends an expanded investigation of long-term watertable adjustment, possibly followed by removal of some floodplain infrastructure or designs to enhance watertable tolerance.

Keywords: Urban restoration; Rain garden; Low impact development; Watertable recharge; Infiltration; Syracuse; NY; Onondaga Creek

Ülo Mander^a and William J. Mitsch^b. (^aDepartment of Geography, Institute of Ecology and Earth Sciences, University of Tartu, 46 Vanemuise St., Tartu 51014, Estonia, ^bWilma H. Schiermeier Olentangy River Wetland Research Park, The Ohio State University, 352 W. Doridge St., Columbus, OH 43202, USA). **Pollution control by wetlands. Ecological Engineering, Volume 35(2) (2009): 153-158**

The 2nd International Symposium on Wetland Pollutant Dynamics and Control (WETPOL 2007), organised by the Department of Geography of the University of Tartu (Estonia) in co-operation with partners from the Estonian University of Life Sciences (Tartu, Estonia), Ghent University (Belgium), and the UNESCO-IHE (Delft, The Netherlands), was held 16–20 September 2007, in Tartu, Estonia. At this meeting, 140 oral presentations (including 9 keynote speeches) and 70 posters by representatives from 38 countries were presented. About half of the presentations considered purification processes in both semi-natural and constructed wetlands. The editorial paper highlights trends in studying the cycling of nitrogen, phosphorus, carbon, heavy metals, and organic pollutants in wetlands, but also in the modelling of pollutant removal and the functioning of plants in the wetland environment. It also describes the WETPOL 2007 meeting, which served as the source of the selected papers, and briefly explains the main aspects of these papers.

Keywords: Constructed wetlands; Heavy metals; Nitrogen removal; Organic pollutants; Pharmaceuticals; Phosphorus retention; Wetland modelling; Wetland restoration

A.C. Bastos^a and N. Magan^{1, a}. (^aApplied Mycology, Cranfield Health, Cranfield University, Vincent Building, MK43 0AL Bedfordshire, UK). ***Trametes versicolor*: Potential for atrazine bioremediation in calcareous clay soil, under low water availability conditions. International Biodeterioration & Biodegradation, Volume 63(4) (2009): 389-394**

This study examined the feasibility of *Trametes versicolor* to actively degrade atrazine ($0.5 \mu\text{g g}^{-1}$) in non-sterile calcareous clay soil (Algarve, Portugal) microcosms for up to 24 weeks ($20 \text{ }^\circ\text{C}$), under low water availability (soil water potentials of -0.7 and -2.8 MPa). Soil

respiration, laccase activity, and atrazine quantification by high-performance liquid chromatography (HPLC) were assessed. Respiration was significantly ($p < 0.05$) enhanced in soil containing the inoculant, particularly in the presence of atrazine, indicating that it remained metabolically active throughout the study. Furthermore, up to 98% and 85% (at -0.7 and -2.8 MPa, respectively) of atrazine was degraded in soil containing both the atrazine and the inoculant, compared to 96% and 50% in soil containing atrazine only. The contribution of *T. versicolor* to atrazine degradation was only significant ($p < 0.005$) under the driest soil treatment conditions. The strategies used for enhancing colonisation and biodegradation capabilities of the inoculant, as well as the selection of sawdust as carrier, were thus effective. However, there were no differences ($p > 0.05$) in quantified laccase activity in soil containing the inoculant and the control. Overall, this study demonstrated that *T. versicolor* was a strong candidate for atrazine bioremediation in soil with low moisture and organic matter contents, such as that found in semi-arid and Mediterranean-like ecosystems.

Keywords: *Trametes versicolor*; Biodegradation; Atrazine; Soil microcosms; Water potential; Soil respiration; Laccase activity

Bella Devassy Tony^b, Dinesh Goyal^b and Sunil Khanna^a. (^aDepartment of Biotechnology and Bioinformatics, NIIT Institute of Information Technology, Balaji Estate, Kalkaji, New Delhi 110019, India, ^bDepartment of Biotechnology and Environmental Sciences, Thapar University, Patiala 147004, Punjab, India). **Decolorization of textile azo dyes by aerobic bacterial consortium. International Biodeterioration & Biodegradation, Volume 63(4) (2009): 462-469**

The decolorization potential of two bacterial consortia developed from a textile wastewater treatment plant showed that among the two mixed bacterial culture SKB-II was the most efficient in decolorizing individual as well as mixture of dyes. At 1.3 g L^{-1} starch supplementation in the basal medium by the end of 120 h decolorization of 80–96% of four out of the six individual azo dyes Congo red, Bordeaux, Ranocid Fast Blue and Blue BCC (10 mg L^{-1}) was noted. The culture exhibited good potential ability in decolorizing 50–60% of all the dyes (Congo red, Bordeaux, Ranocid Fast Blue and Blue BCC) when present as a mixture at 10 mg L^{-1} . The consortium SKB-II consisted of five different bacterial types identified by 16S rDNA sequence alignment as *Bacillus vallismortis*, *Bacillus pumilus*, *Bacillus cereus*, *Bacillus subtilis* and *Bacillus megaterium* which were further tested to decolorize dyes. The efficient ability of this developed consortium SKB-II to decolorize individual dyes and textile effluent using packed bed reactors is being carried out.

Keywords: Azo dyes; Bacterial consortium; Carbon source; Decolorization; *Bacillus vallismortis*; *Bacillus megaterium*

Miguel J.L. Lourenço^a and José Paulo Sampaio^{1, a}. (^aCentro de Recursos Microbiológicos, Departamento de Ciências da Vida, Faculdade de Ciências e Tecnologia, Universidade Nova de Lisboa, Quinta da Torre, 2829-516 Caparica, Portugal). **Microbial deterioration of gelatin emulsion photographs: Differences of susceptibility between black and white and colour materials. International Biodeterioration & Biodegradation, Volume 63(4) (2009): 496-502**

Microbial deterioration is a common problem in photographic collections, and has been considered a major cause of deterioration. However, few studies have been carried out on this topic, and most of the literature concerns biodeterioration of archival documents in general, including both micro- and macroorganisms. There have been no detailed studies on the interactions between microorganisms, environment, and the composition of photographic material. This study focuses on fungal deterioration of gelatin emulsion photographs. It was part of a study of three collections in Lisbon, Portugal. The first part is quantitative research on the fungal contamination of the Horácio Novais collection, and the second involves induced contamination of experiments on gelatin emulsion photographs. At the end these data are analysed, taking into account the hypothesis that colour materials are more susceptible to fungal deterioration than are black and white ones. This hypothesis is based on the observations of professionals working with photograph collections who report that, at least in plastic base supports (negatives and slides), colour materials are frequently more contaminated than the black and white ones. An overall look at the results seems to indicate a higher susceptibility of the colour chromogenic photographic materials to fungal colonization compared to the black and white materials. However, this hypothesis could not be absolutely confirmed by this study.

Keywords: Photographs; Gelatin emulsion; Fungi; Induced contamination

Pensri Plangklang^{a, b} and Alissara Reungsang^{b, c, d}. (^aInternational Postgraduate Programs in Environmental Management, Graduate School, Chulalongkorn University, Bangkok 10330, Thailand, ^bNational Center of Excellence for Environmental and Hazardous Waste Management (NCE-EHWM), Chulalongkorn University, Bangkok 10330, Thailand, ^cDepartment of Biotechnology, Faculty of Technology, Khon Kaen University, Khon Kaen 40002, Thailand, ^dResearch Centre for Environmental and Hazardous Substance Management, Khon Kaen University, Khon Kaen 40002, Thailand). Bioaugmentation of carbofuran residues in soil using *Burkholderia cepacia* PCL3 adsorbed on agricultural residues. *International Biodeterioration & Biodegradation*, Volume 63(4) (2009): 515-522

Burkholderia cepacia PCL3 (GenBank accession number of EF990634) is a carbofuran degrader isolated from phytoremediated rhizosphere soil in our laboratory. Free and the immobilized PCL3 on corncob and sugarcane bagasse were investigated for their abilities to degrade carbofuran in Basal Salt Medium (BSM) and soil microcosm. The reusability and survival of immobilized PCL3 in comparison to free cells were also examined. Short half-lives ($t_{1/2}$) of carbofuran of 3–4 d in BSM were obtained using the isolate PCL3 in both free and immobilized cell forms. Immobilized cells could survive (10^6 – 10^7 cfu ml⁻¹) through 30 d of incubation, while the number of free cells decreased continuously after 10 d. Immobilized *B. cepacia* PCL3 could be reused twice without loss in their abilities to degrade carbofuran in BSM, which suggested an advantage of using immobilized cell over free cell. Free and immobilized cells were augmented into soil and showed an effective capability to remediate carbofuran residues, both of which indicated by 5-folds decrease in carbofuran half-lives in augmented soil. Immobilization of PCL3 on corncob and sugarcane bagasse provided the possibilities of reusing the cells as well as improving the cell survival without decreasing carbofuran degradation activity.

Keywords: Bioaugmentation; *Burkholderia cepacia* PCL3; Carbofuran; Immobilization

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China). Biological treatment of oilfield-produced water: A field pilot study. International Biodeterioration & Biodegradation, Volume 63(3) (2009): 316-321

A field test was conducted on a hydrolysis acidification/bio-contact oxidation system (HA/BCO) to treat oilfield-produced water with high salinity. By operating the biodegradation system for three months with a hydraulic retention time (HRT) of 32 h and a volumetric load of 0.28 kg COD m³ d⁻¹, the treatment process achieved mean removal efficiencies of 63.5% for chemical oxygen demand (COD), 45% for NH₃-N, 79.5% for total suspended solid (TSS), and 68.0% for total petroleum hydrocarbon (TPH). GC/MS was used to analyze relative changes of components of main organic waste in a process indicating that the influent wastewater contained organic compounds from C₁₂H₂₆ to C₃₅H₇₂, which could be degraded effectively with the coordinated action of hydrolysis acidification and aerobic treatment. The use of maize powder can enhance environmental adaptability of microorganisms and biodegradation ability and is recommended as a nutrient supplement to maintain good treatment performance.

Keywords: Oilfield-produced water; Biological treatment; Total petroleum hydrocarbon; Biodegradation; GC/MS

I. Douskova¹, J. Doucha¹, K. Livansky¹, J. Machat², P. Novak³, D. Umysova¹, V. Zachleder¹ and M. Vitova¹. (¹Laboratory of Cell Cycles of Algae, Department of Autotrophic Microorganisms, Institute of Microbiology, Academy of Sciences of the Czech Republic, Novohradská 237, Opatovický mlyn, 379 81 Trebon, Czech Republic, ²Research Centre for Environmental Chemistry and Ecotoxicology, Masaryk University, Kamenice 126/3, 62500 Brno, Czech Republic, ³Termizo Inc., Dr. M. Horakove 571/56, 46006 Liberec, Czech Republic). **Simultaneous flue gas bioremediation and reduction of microalgal biomass production costs. Applied Microbiology and Biotechnology, Volume 82(1) (2009): 179-185**

A flue gas originating from a municipal waste incinerator was used as a source of CO₂ for the cultivation of the microalga *Chlorella vulgaris*, in order to decrease the biomass production costs and to bioremediate CO₂ simultaneously. The utilization of the flue gas containing 10–13% (v/v) CO₂ and 8–10% (v/v) O₂ for the photobioreactor agitation and CO₂ supply was proven to be convenient. The growth rate of algal cultures on the flue gas was even higher when compared with the control culture supplied by a mixture of pure CO₂ and air (11% (v/v) CO₂). Correspondingly, the CO₂ fixation rate was also higher when using the flue gas (4.4 g CO₂ l⁻¹ 24 h⁻¹) than using the control gas (3.0 g CO₂ l⁻¹ 24 h⁻¹). The toxicological analysis of the biomass produced using untreated flue gas showed only a slight excess of mercury while all the other compounds (other heavy metals, polycyclic aromatic hydrocarbons, polychlorinated dibenzodioxins and dibenzofurans, and polychlorinated biphenyls) were below the limits required by the European Union foodstuff legislation. Fortunately, extending the flue gas treatment prior to the cultivation unit by a simple granulated activated carbon column led to an efficient absorption of gaseous mercury and to the algal biomass composition compliant with all the foodstuff legislation requirements.

Keywords: Algae - *Chlorella* - Carbon dioxide - Bioremediation - Flue gas - Food and Feed supply

Nadja Kabelitz¹, Jirina Machackova², GwenaËl Imfeld³, Maria Brennerova⁴, Dietmar H. Pieper⁵, Hermann J. Heipieper¹ and Howard Junca⁵. (¹Department of Bioremediation, Helmholtz Centre for Environmental Research (UFZ), Permoserstr. 15, 04318 Leipzig, Germany, ²Earth Tech CZ s.r.o., TrojskÁ; 92, 171 00 Prague 7, Czech Republic, ³Department of Isotope Biogeochemistry, Helmholtz Centre for Environmental Research (UFZ), Permoserstr. 15, 04318 Leipzig, Germany, ⁴Institute of Microbiology (IMIC), Czech Academy of Sciences, Videnska 1083, 142 20 Prague 4-Krc, Czech Republic, ⁵Biodegradation Research Group, Helmholtz Centre for Infection Research (HZI), Inhoffenstrasse 7, 38124 Braunschweig, Germany). **Enhancement of the microbial community biomass and diversity during air sparging bioremediation of a soil highly contaminated with kerosene and BTEX. *Applied Microbiology and Biotechnology*, Volume 82 (3) (2009): 565-577**

In order to obtain insights in complexity shifts taking place in natural microbial communities under strong selective pressure, soils from a former air force base in the Czech Republic, highly contaminated with jet fuel and at different stages of a bioremediation air sparging treatment, were analyzed. By tracking phospholipid fatty acids and 16S rRNA genes, a detailed monitoring of the changes in quantities and composition of the microbial communities developed at different stages of the bioventing treatment progress was performed. Depending on the length of the air sparging treatment that led to a significant reduction in the contamination level, we observed a clear shift in the soil microbial community being dominated by Pseudomonads under the harsh conditions of high aromatic contamination to a status of low aromatic concentrations, increased biomass content, and a complex composition with diverse bacterial taxonomical branches.

Keywords: BTEX - Air sparging - Bioremediation - Biodiversity - Microbiota

Igwe, J. C.^{1*}, Ekwuruke, A.¹, Gbaruko, B. C.² and Abia, A. A.³. (¹Department of Industrial Chemistry, Abia State University, P.M.B. 2000 Uturu, Abia State, Nigeria, ²Department of Chemistry, Jackson State University, Jackson, MS 39217, U.S.A., ³Department of Pure and Industrial Chemistry, University of Port Harcourt, River State, Nigeria., *Corresponding author email: jcgwe2001@yahoo.com). **Detoxification of copper fungicide using EDTA-modified cellulosic material. *African Journal of Biotechnology* Volume 8 (3) (2009): 499–506**

Pesticides are poisons and can be particularly dangerous when misused or carelessly disposed. The detoxification of a copper fungicide (KOCIDE 101) using maize cob, a cellulosic material, was studied. Based on copper as the active agent (after a sorption period of 1 h), the concentration of the fungicide reduced from an initial value of 2000 to 206.25 ppm for the unmodified maize cob and to 24.31 ppm for the modified maize cob. The pseudo-first and second order rate equations were used to model the detoxification process. The intraparticle diffusivity and mechanism of the sorption was proposed. Also, equilibrium sorption isotherms were evaluated using the Freundlich, Langmuir and Dubinin-Radushkevich isotherm models. This results show that maize cob is an effective adsorbent for copper fungicide deactivation and detoxification.

Key words: Copper fungicide, maize cob, adsorption, mechanism, EDTA, detoxification

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Marine Chemicals Research Institute, Bhavnagar, 364 002, Gujarat, India). Biosorption of Cd(II) and Pb(II) onto brown seaweed, *Lobophora variegata* (Lamouroux): kinetic and equilibrium studies Volume 20(1) (2009): 1-13

The present work deals with the biosorption performance of raw and chemically modified biomass of the brown seaweed *Lobophora variegata* for removal of Cd(II) and Pb(II) from aqueous solution. The biosorption capacity was significantly altered by pH of the solution delineating that the higher the pH, the higher the Cd(II) and Pb(II) removal. Kinetic and isotherm experiments were carried out at the optimal pH 5.0. The metal removal rates were conspicuously rapid wherein 90% of the total sorption occurred within 90 min. Biomass treated with CaCl₂ demonstrated the highest potential for the sorption of the metal ions with the maximum uptake capacities i.e. 1.71 and 1.79 mmol g⁻¹ for Cd(II) and Pb(II), respectively. Kinetic data were satisfactorily manifested by a pseudo-second order chemical sorption process. The process mechanism consisting of both surface adsorption and pore diffusion was found to be complex. The sorption data have been analyzed and fitted to sorption isotherm of the Freundlich, Langmuir, and Redlich–Peterson models. The regression coefficient for both Langmuir and Redlich–Peterson isotherms were higher than those secured for Freundlich isotherm implying that the biosorption system is possibly monolayer coverage of the *L. variegata* surface by the cadmium and lead ions. FT-IR studies revealed that Cd(II) and Pb(II) binding to *L. variegata* occurred primarily through biomass carboxyl groups accompanied by momentous interactions of the biomass amino and amide groups. In this study, we have observed that *L. variegata* had maximum biosorption capacity for Cd(II) and Pb(II) reported so far for any marine algae.

Keywords: Brown seaweed - *Lobophora variegata* - Biosorption - Kinetics - Isotherm - FT-IR

V. Faraco^{1, 2}, C. Pezzella¹, A. Miele¹, P. Giardina¹ and G. Sannia¹. (¹Department of Organic Chemistry and Biochemistry, University of Naples “Federico II”, Complesso Universitario Monte S. Angelo, via Cintia, 4, 80126 Naples, Italy, ²School of Biotechnological Sciences, University of Naples “Federico II”, 80126 Naples, Italy). Bioremediation of colored industrial wastewaters by the white-rot fungi *Phanerochaete chrysosporium* and *Pleurotus ostreatus* and their enzymes. *Biodegradation*, Volume 20(2) (2009): 209-220

The effect of *Phanerochaete chrysosporium* and *Pleurotus ostreatus* whole cells and their ligninolytic enzymes on models of colored industrial wastewaters was evaluated. Models of acid, direct and reactive dye wastewaters from textile industry have been defined on the basis of discharged amounts, economic relevance and representativeness of chemical structures of the contained dyes. *Phanerochaete chrysosporium* provided an effective decolourization of direct dye wastewater model, reaching about 45% decolourization in only 1 day of treatment, and about 90% decolourization within 7 days, whilst *P. ostreatus* was able to decolorize and detoxify acid dye wastewater model providing 40% decolourization in only 1 day, and 60% in 7 days. *P. ostreatus* growth conditions that induce laccase production (up to 130,000 U/l) were identified, and extra-cellular enzyme mixtures, with known laccase isoenzyme composition, were produced and used in wastewater models decolourization. The mixtures decolorized and detoxified the acid dye wastewater model, suggesting laccases as the main agents of wastewater decolourization by *P. ostreatus*. A laccase mixture was immobilized by entrapment in Cu-

alginate beads, and the immobilized enzymes were shown to be effective in batch decolourization, even after 15 stepwise additions of dye for a total exposure of about 1 month.

Keywords: Textile dyes decolourization - Industrial effluent treatment - Ligninolytic fungi - Laccase - Manganese-peroxidase - Color industry

Tong-Jiang Xu^a and Yen-Peng Ting^a. (^aDepartment of Chemical and Biomolecular Engineering, National University of Singapore, 4 Engineering Drive 4, Singapore 117576, Singapore). **Fungal bioleaching of incineration fly ash: Metal extraction and modeling growth kinetics. Enzyme and Microbial Technology, Volume 44(5) (2009): 323-328**

Aspergillus niger is known to be capable of bioleaching heavy metal ions from municipal solid waste (MSW) incineration fly ash. The objective of this study was to investigate the bioleaching kinetics of the fungus in the presence of the fly ash at various pulp densities (1–6%) in a batch system. The growth of the fungus was modeled using the modified Gompertz model. Since the metals present in the fly ash are toxic and inhibit microbial growth, an inhibition kinetic model using the generalized Monod growth kinetics was evaluated. In a two-step bioleaching system where the fly ash was introduced into the culture two days after inoculation, citric acid production and the leaching of the metals aluminium, iron and zinc from the fly ash were examined. The kinetic parameters in the system were estimated using the least square method. Results showed that the modified Gompertz model fit the experimental data well. The specific growth rate decreased with increasing pulp density, with a maximum specific growth rate (μ_{\max}) of 0.115 day^{-1} for the control. The critical inhibitor (i.e. fly ash) concentration (C_i) above which no growth occurred was found to be 6.0%. Results also showed an increase in metal concentration leached with a concomitant increase in the citric acid production at various pulp densities.

Keywords: *Aspergillus niger*; Bioleaching; Incineration fly ash; Gompertz model; Han and Levenspiel model; Inhibition

Lei Yao^a, Zheng-fang Ye^a, Mei-ping Tong^a, Peng Lai^a and Jin-ren Ni^a. (^aDepartment of Environmental Engineering, Peking University, The Key Laboratory of Water and Sediment Sciences, Ministry of Education, Beijing 100871, China). **Removal of Cr^{3+} from aqueous solution by biosorption with aerobic granules. Journal of Hazardous Materials, Volume 165(1-3) (2009): 250-255**

Aerobic granules were utilized as an effective biosorbent to remove Cr^{3+} from aqueous solution. The results showed that the initial pH, contact time, and Cr^{3+} concentration affected the biosorption process significantly. Both Freundlich and Langmuir isotherms were able to describe the equilibrium data reasonably with high correlation coefficients ($R^2 > 0.95$) and pseudo-second-order model best fitted the biosorption process at experimental conditions. Moreover, Environmental Scanning Electronic microscope (ESEM), X-ray energy dispersion (EDX), and Fourier transform infrared (FTIR) analyses revealed that metal complexation, chemical precipitation, and ion exchange were involved in the removal of Cr^{3+} with aerobic granules. Further analysis by a metal ion fraction test demonstrated that metal complexation could be the dominant mechanism of biosorption, whereas chemical precipitation and ion exchange appeared only to have minor role in the overall Cr^{3+} biosorption process.

Keywords: Biosorption; Cr^{3+} ; Aerobic granules; ESEM; Metal ion fraction

Mustafa Tuzen^a, Ahmet Sari^a, Durali Mendil^a, Ozgur Dogan Uluozlu^a, Mustafa Soylak^b and Mehmet Dogan^c. (^aDepartment of Chemistry, Gaziosmanpasa University, 60250, Tokat, Turkey, ^bDepartment of Chemistry, Erciyes University, 38039, kayseri, Turkey, ^cDepartment of Chemistry, Hacettepe University, Ankara, Turkey). **Characterization of biosorption process of As(III) on green algae *Ulothrix cylindricum*. Journal of Hazardous Materials, Volume 165(1-3) (2009): 566-572**

Arsenic (As) is generally found as As(III) and As(V) in environmental samples. Toxicity of As(III) is higher than As(V). This paper presents the characteristics of As(III) biosorption from aqueous solution using the green algae (*Ulothrix cylindricum*) biomass as a function of pH, biomass dosage, contact time, and temperature. Langmuir, Freundlich and Dubinin–Radushkevich (D–R) models were applied to describe the biosorption isotherm of As(III) by *U. cylindricum* biomass. The biosorption capacity of *U. cylindricum* biomass was found as 67.2 mg/g. The metal ions were desorbed from *U. cylindricum* using 1 M HCl. The high stability of *U. cylindricum* permitted 10 times of adsorption–elution process along the studies with a slightly decrease about 16% in recovery of As(III) ions. The mean free energy value evaluated from the D–R model indicated that the biosorption of As(III) onto *U. cylindricum* biomass was taken place by chemical ion-exchange. The calculated thermodynamic parameters, ΔG° , ΔH° and ΔS° showed that the biosorption of As(III) onto *U. cylindricum* biomass was feasible, spontaneous and exothermic under examined conditions. Experimental data were also tested in terms of biosorption kinetics using pseudo-first-order and pseudo-second-order kinetic models. The results showed that the biosorption processes of As(III) followed well pseudo-second-order kinetics.

Keywords: *U. Cylindricum*; Green algae; Biosorption; As(III)

Kuber C. Bhainsa^a and Stanislaus F. D'Souza^a. (^aNuclear Agriculture and Biotechnology Division, Bhabha Atomic Research Centre, Trombay, Mumbai 400085, India). **Thorium biosorption by *Aspergillus fumigatus*, a filamentous fungal biomass. Journal of Hazardous Materials, Volume 165(1-3) (2009): 670-676**

Thorium biosorption by *Aspergillus fumigatus* was carried out in a batch reactor to study the effect of initial pH and metal ion concentration, contact time, biomass dose and kinetics and equilibrium Th uptake. Thorium(IV) uptake by *A. fumigatus* was pH dependent (pH range, 2.0–6.0) and maximum sorption was observed at pH 4.0. The uptake was rapid and the biosorption process reached equilibrium within 2 h of contact times at pH 2–4 and initial Th concentration of 50 and 100 mg/L. The kinetics data fitted well to Lagergren's pseudo-second-order rate equation ($r^2 > 0.99$). A maximum initial sorption rate of 71.94 (mg/g min) and second-order rate constant of 7.82×10^{-2} (g/mg min) were observed at pH 4.0, 50 mg Th/L. The observed maximum uptake of thorium was 370 mg Th/g at equilibrium. Biosorption process could be well described by Langmuir isotherm in comparison to Freundlich and Temkin isotherms. Sodium bicarbonate was the most efficient desorbing reagent with desorption efficiency of more than 99%. Environmental scanning electron micrograph (ESEM) showed that the surface of the biomass after desorption was intact.

Keywords: Thorium; *Aspergillus fumigatus*; Biosorption; Isotherm; Desorption

Hai-ping Yuan^a, Jun-hui Zhang^{a, b}, Zhen-mei Lu^a, Hang Min^a and Chu Wu^c. (^aCollege of Life Science, Zhejiang University, 310058 Hangzhou, PR China, ^bCollege of Life Science, Taizhou University, 317000 Taizhou, PR China, ^cSchool of Life & Environmental Sciences, Wenzhou University, 325027 Wenzhou, PR China). **Studies on biosorption equilibrium and kinetics of Cd²⁺ by *Streptomyces* sp. K33 and HL-12. *Journal of Hazardous Materials*, Volume 164(2-3) (2009): 423-431**

The sorption of Cd²⁺ by *Streptomyces* sp. K33 and HL-12 was investigated. The removal efficiency increased with pH, but no obvious differences with different temperatures. Fourier transform infrared (FT-IR) was used to characterize the interaction between Cd²⁺ and K33 and HL-12. Results revealed that the presence of amino, carboxyl, hydroxyl and carbonyl groups were responsible for the biosorption of Cd²⁺. Strain HL-12 had more changes in the functional groups than K33. Biosorption equilibrium was established earlier by strain K33 than that by HL-12, and K33 had higher adsorption ratio. Langmuir, Freundlich and Dubinin–Radushkevich (D–R) isotherms were used to describe the adsorption experiment, Langmuir model fitted the experiment data best. Strain K33 showed greater sorption capacities with 38.49 mg Cd²⁺/g dry cells. Pseudo-first-order and second-order kinetic models were used to describe the kinetic data, and second-order kinetic model fitted better. About 70% recovery of Cd²⁺ could be obtained at pH ≤ 3 from metal-loaded biomass of strains HL-12 and K33.

Keywords: *Streptomyces* sp.; Biosorption; Cadmium; Desorption; FT-IR

Erkan Sahinkaya^a. (^aHarran University, Environmental Engineering Department, Osmanbey Campus, 63000 Sanliurfa, Turkey). **Biotreatment of zinc-containing wastewater in a sulfidogenic CSTR: Performance and artificial neural network (ANN) modelling studies. *Journal of Hazardous Materials*, Volume 164(1) (2009): 105-113**

Sulfidogenic treatment of sulfate (2–10 g/L) and zinc (65–677 mg/L) containing simulated wastewater was studied in a mesophilic (35 °C) CSTR. Ethanol was supplemented (COD/sulfate = 0.67) as carbon and energy source for sulfate-reducing bacteria (SRB). The robustness of the system was studied by increasing Zn, COD and sulfate loadings. Sulfate removal efficiency, which was 70% at 2 g/L feed sulfate concentration, steadily decreased with increasing feed sulfate concentration and reached 40% at 10 g/L. Over 99% Zn removal was attained due to the formation of zinc-sulfide precipitate. COD removal efficiency at 2 g/L feed sulfate concentration was over 94%, whereas, it steadily decreased due to the accumulation of acetate at higher loadings. Alkalinity produced from acetate oxidation increased wastewater pH remarkably when feed sulfate concentration was 5 g/L or lower. Electron flow from carbon oxidation to sulfate reduction averaged 83 ± 13%. The rest of the electrons were most likely coupled with fermentative reactions as the amount of methane production was insignificant. The developed ANN model was very successful as an excellent to reasonable match was obtained between the measured and the predicted concentrations of sulfate ($R = 0.998$), COD ($R = 0.993$), acetate ($R = 0.976$) and zinc ($R = 0.827$) in the CSTR effluent.

Keywords: CSTR; Sulfate reduction; Metal removal; Zinc removal; Artificial neural network

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Serbia). Biosorption of heavy metal ions from aqueous solutions by short hemp fibers: Effect of chemical composition. Journal of Hazardous Materials, Volume 164(1) (2009): 146-153

Sorption potential of waste short hemp fibers for Pb^{2+} , Cd^{2+} and Zn^{2+} ions from aqueous media was explored. In order to assess the influence of hemp fiber chemical composition on their heavy metals sorption potential, lignin and hemicelluloses were removed selectively by chemical modification. The degree of fiber swelling and water retention value were determined in order to evaluate the change in accessibility of the cell wall components to aqueous solutions due to the fiber modification. The effects of initial ion concentration, contact time and cosorption were studied in batch sorption experiments. The obtained results show that when the content of either lignin or hemicelluloses is progressively reduced by chemical treatment, the sorption properties of hemp fibers are improved. Short hemp fibers are capable of sorbing metal ions (Pb^{2+} , Cd^{2+} and Zn^{2+}) from single as well as from ternary metal ion solutions. The maximum total uptake capacities for Pb^{2+} , Cd^{2+} and Zn^{2+} ions from single solutions are the same, i.e. 0.078 mmol/g, and from ternary mixture 0.074, 0.035 and 0.035 mmol/g, respectively.

Keywords: Short hemp fibers; Chemical modification; Biosorption; Heavy metals; Fiber swelling

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Environmental contamination caused by radionuclides, in particular by uranium and its decay products is a serious problem worldwide. The development of nuclear science and technology has led to increasing nuclear waste containing uranium being released and disposed in the environment.

The objective of this paper is to develop a better understanding of the techniques for the remediation of soils polluted with radionuclides (uranium in particular), considering: the chemical forms of uranium, including depleted uranium (DU) in soil and other environmental media, their characteristics and concentrations, and some of the effects on environmental and human health; research issues concerning the remediation process, the benefits and results; a better understanding of the range of uses and situations for which each is most appropriate.

The paper addresses the main features of the following techniques for uranium remediation: natural attenuation, physical methods, chemical processes (chemical extraction methods from contaminated soils assisted by various suitable chelators (sodium bicarbonate, citric acid, two-stage acid leaching procedure), extraction using supercritical fluids such as solvents, permeable reactive barriers), biological processes (biomineralization and microbial reduction, phytoremediation, biosorption), and electrokinetic methods. In addition, factors affecting uranium removal from soils are furthermore reviewed including soil characteristics, pH and reagent concentration, retention time.

Keywords: Bioremediation; Biosorption; Bioreduction; Chemical extraction; Electrokinetics; Environment; Polluted soil; Remediation; Uranium

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The physicochemical characteristics of three Brazilian peats were investigated using elemental analysis, scanning electron microscopy (SEM), X-ray diffractometry (XRD) and studies of Cr(III) biosorption based on adsorption isotherms. Adsorption of Cr(III) by *in natura* peat from Santo Amaro das Brotas (Sergipe State) was much greater than by peats from either Ribeirão Preto (São Paulo State) or Itabaiana (Sergipe State), with adsorption capacities (q) of 4.90 ± 0.01 , 1.70 ± 0.01 and 1.40 ± 0.01 mg g⁻¹, respectively. Pre-treatments with HCl and NaOH + HCl reduced adsorption by the Santo Amaro das Brotas peat, showing that adsorption efficiency was associated with the amount of organic matter present. Conversely, increase in the mineral content following pre-treatment increased adsorption of Cr(III) by the Ribeirão Preto and Itabaiana peats. Highest adsorption (retention >95.0%) was achieved at equilibrium pH 4.0 using the Santo Amaro das Brotas peat. Experimental data for the adsorption of Cr(III) from aqueous solution onto this peat were fitted to the Langmuir equation, from which an equilibrium adsorption capacity, q_{max} , of 5.60 mg g⁻¹ was obtained, which was close to the experimentally determined value.

Keywords: Adsorption; Chromium; Peat; Chemical pre-treatment

Y.N. Mata^a, M.L. Blázquez^a, A. Ballester^a, F. González^a and J.A. Muñoz^a. (^aDepartment of Materials Science and Metallurgical Engineering, Complutense University of Madrid, Ciudad Universitaria s/n, 28040 Madrid, Spain). **Biosorption of cadmium, lead and copper with calcium alginate xerogels and immobilized *Fucus vesiculosus*. Journal of Hazardous Materials, Volume 163(2-3) (2009): 555-562**

This paper determines the effect of immobilized brown alga *Fucus vesiculosus* in the biosorption of heavy metals with alginate xerogels. Immobilization increased the kinetic uptakes and intraparticle diffusion rates of the three metals. The Langmuir maximum biosorption capacity increased twofold for cadmium, 10 times for lead, and decreased by half for copper. According to this model, the affinity of the metals for the biomass was as follows: Cu > Pb > Cd without alga and Pb > Cu > Cd with alga. FITR confirmed that carboxyl groups were the main groups involved in the metal uptake. Calcium in the gels was displaced by heavy metals from solution according to the “egg-box” model. The restructured gel matrix became more uniform and organized as shown by scanning electron microscopy (SEM) characterization. *F. vesiculosus* immobilized in alginate xerogels constitutes an excellent biosorbent for cadmium, lead and copper, sometimes surpassing the biosorption performance of alginate alone and even the free alga.

Keywords: Biosorption; Heavy metals; Alginate; Immobilization; *Fucus vesiculosus*

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In this paper, marine brown algae *Laminaria japonica* was chemically modified by crosslinking with epichlorohydrin (EC₁ and EC₂), or oxidizing by potassium permanganate (PC), or crosslinking with glutaraldehyde (GA), or only washed by distilled water (DW). They were used for equilibrium sorption uptake studies with Cd²⁺, Cu²⁺, Ni²⁺ and Zn²⁺. The experimental data have been analyzed using Langmuir, Freundlich and Redlich–Peterson isotherms. The results showed that the biosorption equilibrium was well described by both the Langmuir and Redlich–Peterson isotherms. The order of maximum metal uptakes for Cd²⁺, Cu²⁺ and Zn²⁺ was EC₁ > EC₂ > PC > DW > GA, but the uptakes of Ni²⁺ are almost the same for these sorbents. Moreover, sorption kinetics has been performed and it was observed that the equilibrium was reached in less than 2 h, which could be described by pseudo-first-order kinetic model. The metal adsorption was strictly pH dependent. The optimum pH values of four metals were in the range of 4.3–6.5 for all sorbents, and the optimum solid/liquid ratio was 3.0 g L⁻¹.

Keywords: Algae; Biosorption; Heavy metal; Chemical modification

Ruofei Jin^a, Hua Yang^a, Aili Zhang^a, Jing Wang^a and Guangfei Liu^a. (^aSchool of Environmental and Biological Science and Technology, Dalian University of Technology, Dalian 116023, PR China). Bioaugmentation on decolorization of C.I. Direct Blue 71 by using genetically engineered strain *Escherichia coli* JM109 (pGEX-AZR). *Journal of Hazardous Materials*, Volume 163(2-3) (2009): 123-1128

The study showed that *Escherichia coli* JM109 (pGEX-AZR), the genetically engineered microorganism (GEM) with higher ability to decolorize azo dyes, bioaugmented successfully the dye wastewater bio-treatment systems to enhance C.I. Direct Blue 71 (DB 71) decolorization. The control and bioaugmented reactors failed at a around pH 5.0. However, the bioaugmented one succeeded at around pH 9.0, the influent DB 71 concentration was 150 mg/L, DB 71 concentration was decreased to 27.4 mg/L in 12 h. The 1–3% NaCl concentration of bioaugmented reactors had no definite influence on decolorization, DB 71 concentration was decreased to 12.6 mg/L in 12 h. GEM was added into anaerobic sequencing batch reactors (AnSBRs) to enhance DB 71 decolorization. Continuous operations of the control and bioaugmented AnSBRs showed that *E. coli* JM109 (pGEX-AZR) could bioaugment decolorization. The concentrations of activated sludge and GEM were still more than 2.80 g/L and 1.5 × 10⁶ cells/mL, respectively, in the bioaugmented AnSBR. All the microbial communities changed indistinctively with time. The microbial community structures of the control AnSBR were similar to those of the bioaugmented one.

Keywords: *Escherichia coli* JM109 (pGEX-AZR); Azo dye; ARDRA; Bioaugmentation; Decolorization

Tamer Akar^a, Zerrin Kaynak^a, Sefika Ulusoy^a, Dilek Yuvaci^a, Guldem Ozsari^a and Sibel Tunali Akar^a. (^aDepartment of Chemistry, Faculty of Arts and Science, Eskişehir Osmangazi University, Campus of Meselik, 26480 Eskişehir, Turkey). **Enhanced biosorption of nickel(II) ions by silica-gel-immobilized waste biomass: Biosorption characteristics in batch and dynamic flow mode. Journal of Hazardous Materials, Volume 163(2-3) (2009): 1134-1141**

Batch and dynamic flow biosorption studies were carried out using the waste biomass entrapped in silica-gel matrix for the removal of nickel(II) ions from synthetic solutions and real wastewater. Batch biosorption conditions were examined with respect to initial pH, S/L ratio, contact time, and initial nickel ion concentration. Zeta potential measurements showed that immobilized biosorbent was negatively charged in the pH range of 3.0–8.0. The immobilized biomass was found to possess relatively high biosorption capacity (98.01 mg g⁻¹), and biosorption equilibrium was established in a short time of operation (5 min). The equilibrium data were followed by Langmuir, Freundlich, and Dubinin–Radushkevich isotherm models. Scanning electron microscope analysis was used to screen the changes on the surface structure of the waste biomass after immobilization and nickel(II) biosorption. Sorbent–sorbate interactions were confirmed by Fourier transform infrared spectroscopy. The applicability of sorbent system was investigated in a continuous mode, and column studies were performed under different flow rate, column size, and biosorbent dosage. Also, the proposed sorbent system was successfully used to remove the nickel ions from industrial wastewater in dynamic flow treatment mode. The results showed that silica-immobilized waste biomass was a low-cost promising sorbent for sequester of nickel(II) ions from synthetic and real wastewater.

Keywords: Biosorption; Nickel; Waste biomass; Immobilization; Real wastewater

Meral Yurtsever^a and İ. Ayhan Şengil^a. (^aDepartment of Environmental Engineering, Engineering Faculty, Sakarya University, 54187 Sakarya, Turkey). **Biosorption of Pb(II) ions by modified quebracho tannin resin. Journal of Hazardous Materials, Volume 163(1) (2009) : 58-64**

In this study, the effect of temperature, pH and initial metal concentration on Pb(II) biosorption on modified quebracho tannin resin (QTR) was investigated. Scanning electron microscopy (SEM) and Fourier transform infrared spectroscopy (FTIR) were used to investigate QTR structure and morphology. Besides, the specific BET surface area and zeta-potential of the QTR were analysed. Thermodynamic functions, the change of free energy (ΔG°), enthalpy (ΔH°) and entropy (ΔS°) of Pb adsorption on modified tannin resin were calculated as $-5.43 \text{ kJ mol}^{-1}$ (at $296 \pm 2 \text{ K}$), $31.84 \text{ kJ mol}^{-1}$ and $0.127 \text{ J mmol}^{-1} \text{ K}^{-1}$, respectively, indicating the spontaneous, endothermic and the increased randomness nature of Pb²⁺ adsorption. The kinetic data was tested using pseudo-first-order, pseudo-second-order, Elovich and intraparticle diffusion model. The results suggested that the pseudo-second-order model ($R^2 > 0.999$) was the best choice among all the kinetic models to describe the adsorption behavior of Pb(II) onto QTR. Langmuir, Freundlich and Tempkin adsorption models were used to represent the equilibrium data. The best interpretation for the experimental data was given by the Langmuir isotherm and the maximum adsorption capacity (86.207 mg g⁻¹) of Pb(II) was obtained at pH 5 and 296 K.

Keywords: Biosorption; Isotherms; Kinetics; Lead; Quebracho tannin resin

Jaeyoung Choi^a, Ju Young Lee^a and Jung-Seok Yang^a. (^aKorea Institute of Science and Technology (KIST), Gangneung Institute, Gangneung 210-340, South Korea). Biosorption of heavy metals and uranium by starfish and *Pseudomonas putida*. *Journal of Hazardous Materials*, Volume 161(1) (2009): 157-162

Biosorption of heavy metals and uranium from contaminated wastewaters may represent an innovative purification process. This study investigates the removal ability of unit mass of *Pseudomonas putida* and starfish for lead, cadmium, and uranium by quantifying the adsorption capacity. The adsorption of heavy metals and uranium by the samples was influenced by pH, and increased with increasing Pb, Cd, and U concentrations. Dead cells adsorbed the largest quantity of all heavy metals than live cells and starfish. The adsorption capacity followed the order: U(VI) > Pb > Cd. The results also suggest that bacterial membrane cells can be used successfully in the treatment of high strength metal-contaminated wastewaters.

Keywords: Adsorption; Heavy metal; *Pseudomonas putida*; Starfish; Uranium

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The ability of white-rot fungus, *Pycnoporus sanguineus* to adsorb copper (II) ions from aqueous solution is investigated in a batch system. The live fungus cells were immobilized into Calcium alginate gel to study the influence of pH, initial metal ions concentration, biomass loading and temperature on the biosorption capacity. The optimum uptake of Cu (II) ions was observed at pH 5 with a value of 2.76 mg/g. Biosorption equilibrium data were best described by Langmuir isotherm model followed by Redlich–Peterson and Freundlich models, respectively. The biosorption kinetics followed the pseudo-second order and intraparticle diffusion equations. The thermodynamic parameters enthalpy change (10.16 kJ/mol) and entropy change (33.78 J/mol K) were determined from the biosorption equilibrium data. The FTIR analysis showed that —OH, —NH, C—H, C=O, —COOH and C—N groups were involved in the biosorption of Cu (II) ions onto immobilized cells of *P. sanguineus*. The immobilized cells of *P. sanguineus* were capable of removing Cu (II) ions from aqueous solution.

Keywords: Biosorption; Copper; *Pycnoporus sanguineus*; Equilibrium; Kinetics

V.K. Gupta^a and A. Rastogi^a. (^aDepartment of Chemistry, Indian Institute of Technology Roorkee, Roorkee 247 667, India). Biosorption of hexavalent chromium by raw and acid-treated green alga *Oedogonium hatei* from aqueous solutions. *Journal of Hazardous Materials*, Volume 163(1) (2009): 396-402

The hexavalent chromium, Cr(VI), biosorption by raw and acid-treated *Oedogonium hatei* were studied from aqueous solutions. Batch experiments were conducted to determine the biosorption properties of the biomass. The optimum conditions of biosorption were found to be: a biomass dose of 0.8 g/L, contact time of 110 min, pH and temperature 2.0 and 318 K respectively. Both Langmuir and Freundlich isotherm equations could fit the equilibrium data. Under the optimal

conditions, the biosorption capacities of the raw and acid-treated algae were 31 and 35.2 mg Cr(VI) per g of dry adsorbent, respectively. Thermodynamic parameters showed that the adsorption of Cr(VI) onto algal biomass was feasible, spontaneous and endothermic under studied conditions. The pseudo-first-order kinetic model adequately describe the kinetic data in comparison to second-order model and the process involving rate-controlling step is much complex involving both boundary layer and intra-particle diffusion processes. The physical and chemical properties of the biosorbent were determined and the nature of biomass–metal ions interactions were evaluated by FTIR analysis, which showed the participation of —COOH, —OH and —NH₂ groups in the biosorption process. Biosorbents could be regenerated using 0.1 M NaOH solution, with up to 75% recovery. Thus, the biomass used in this work proved to be effective materials for the treatment of chromium bearing aqueous solutions.

Keywords: *Oedogonium hatei*; Hexavalent chromium; Biosorption; Thermodynamic parameters; Kinetic models; FTIR

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The biosorption potential of *Racomitrium lanuginosum* as aquatic moss biosorbent for the removal of Pd(II) from aqueous solution was investigated. The effects of pH, biomass dosage, contact time, and temperature on the biosorption processes were systematically studied. Experimental data were modeled by Langmuir, Freundlich and Dubinin–Radushkevich (D–R) isotherms. Langmuir isotherm model ($R^2 = 0.994$) fitted the equilibrium data better than the Freundlich isotherm model ($R^2 = 0.935$). The monolayer biosorption capacity of *R. lanuginosum* biomass for Pd(II) was found to be 37.2 mg/g at pH 5. The mean free energy was calculated as 9.2 kJ/mol using the D–R isotherm model ($R^2 = 0.996$). This result indicated that the biosorption of Pd(II) was taken place by chemical ion-exchange. The calculated thermodynamic parameters, ΔG° , ΔH° and ΔS° showed that the biosorption of Pd(II) on *R. lanuginosum* biomass was feasible, spontaneous and exothermic under examined conditions. Experimental data were also tested using the biosorption kinetic models. The results showed that the biosorption processes of Pd(II) on *R. lanuginosum* followed well pseudo-second-order kinetics at 20–50 °C ($R^2 = 0.999$).

Keywords: *Racomitrium lanuginosum*; Moss; Palladium(II); Biosorption

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Biosorption has been appearing as a useful alternative to conventional treatment systems for the removal of toxic metals from aqueous stream. The batch removal of chromate anions (CrO₄²⁻) from wastewater under different experimental conditions using a cationic surfactant-modified lichen (*Cladonia rangiformis* (L.)) was investigated in this study. Cetyl trimethyl ammonium bromide (CTAB) was used for biomass modification. The results of the experiments showed that

biomass modification substantially improved the biosorption efficiency. Effects of pH, biosorption time, initial CrO_4^{2-} concentration, biosorbent dosage, and the existence of the surfactant on the biosorption of CrO_4^{2-} anions were studied. Studies up to date have shown that the biosorption efficiency of chromium increased as the pH of the solution decreased. In the present study, the removal of chromate anions from aqueous solutions at high pH values with surfactant-modified lichen was investigated. From the results of the experiments it was seen that the removal of chromate anions by modified lichen was 61% at the solution natural pH (pH 5.11) but at the same pH value the removal of chromate anions by unmodified lichen was 6%. Also concentrations ranging from 30 to 150 mg/L Cr(IV) were tested and the biosorptive removal efficiency of the metal ions from aqueous solution at high pH was achieved more than 98%.

Keywords: Biosorption; Chromate anions (CrO_4^{2-}); Surfactant; Lichen

Biotransformation

Lei Cai^{a,1}, Mei-Qing Yuan^{a,b,1}, Feng Liu^a, Jia Jian^a and Guo-Qiang Chen^{a,c}. (^aDepartment of Biological Sciences and Biotechnology, Tsinghua University, Beijing 100084, China, ^bInstitute of Forensic Sciences, Ministry of Public Security, Beijing 100038, China, ^cMultidisciplinary Research Center, Shantou University, Shantou 515063, China). **Enhanced production of medium-chain-length polyhydroxyalkanoates (PHA) by PHA depolymerase knockout mutant of *Pseudomonas putida* KT2442. Bioresource Technology, Volume 100(7) (2009): 2265-2270**

Pseudomonas putida KT2442 is a medium-chain-length polyhydroxyalkanoates (PHA) producer. One of the main shortages in the production of PHA has been the intracellular PHA degradation caused by its endogenous PHA depolymerase. The aim of this study was to improve PHA production via removing the PHA degradation mechanism. PHA depolymerase *phaZ* knockout mutant *P. putida* KTMQ01 was successfully constructed, which accumulated 86 wt% medium-chain-length PHA (mcl PHA) when cultured in mineral medium containing sodium octanoate as the carbon source compared with *P. putida* KT2442 which produced only 66 wt% of its cell dry weight (CDW). *P. putida* KTMQ01 cultured over a five-day period on sodium octanoate produced 4.5 g L^{-1} – 4.0 g L^{-1} CDW containing approximately 80 wt% PHA without degradation. In contrast, *P. putida* KT2442 was observed with decreasing CDW and PHA from over 4 to less than 2 g L^{-1} over the same period of time, indicating the function of PHA depolymerases which reduced the amount of PHA from around 50 wt% to none over the incubation period. RT-PCR analysis showed that *phaC2* transcriptional level of *P. putida* KTMQ01 was higher than that of *P. putida* KT2442, indicating the possibility of relief on negative control of *phaC2* transcription by the deletion of *phaZ*, which combined with the lack of *in vivo* PHA degradation, led to more PHA accumulation. *P. putida* KTMQ01 contained PHA granules with larger sizes and smaller numbers than those of *P. putida* KT2442.

Keywords: PHB; Polyhydroxyalkanoates; *Pseudomonas putida*; Polyhydroxyalkanoates depolymerase; *phaZ*

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This work optimized the novel biotransformation process of podophyllotoxin to produce podophyllic acid by *Pseudomonas aeruginosa* CCTCC AB93066. Firstly, the biotransformation process was significantly affected by medium composition. 5 g/l of yeast extract and 5 g/l of peptone were favorable for podophyllic acid production (i.e. 25.3 ± 3.7 mg/l), while not beneficial for the cell growth of *P. aeruginosa*. This indicated that the accumulation of podophyllic acid was not corresponded well to the cell growth of *P. aeruginosa*. 0 g/l of sucrose was beneficial for podophyllic acid production (i.e. 34.3 ± 3.9 mg/l), which led to high podophyllotoxin conversion (i.e. $98.2 \pm 0.1\%$). 1 g/l of NaCl was the best for podophyllic acid production (i.e. 47.6 ± 4.0 mg/l). Secondly, the production of podophyllic acid was significantly enhanced by fed-batch biotransformation. When each 100 mg/l of podophyllotoxin was added to the biotransformation system after 4, 10 and 25 h of culture, respectively, podophyllic acid concentration reached 99.9 ± 12.3 mg/l, enhanced by 284% comparing to one-time addition (i.e. 26.0 ± 2.1 mg/l). The fundamental information obtained in this study provides a simple and efficient way to produce podophyllic acid.

Keywords: Biotransformation process optimization; Podophyllotoxin; Podophyllic acid; Fed-batch biotransformation; *Pseudomonas aeruginosa*

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The biodegradation of six PCB congeners (IUPAC nos. 28, 52, 101, 138, 153, and 168) present in transformer oil using different commercial mixtures of microorganisms (Sybron 1000, Biozyn 301, Biozyn 300, DBC 5, ChZR, NS 20-10, NS 20-20, and NS 20) under anoxic, oxic, or anoxic/oxic treatments was investigated at laboratory scale. Overall, PCB congener biodegradation was observed in all treatments in the ranges of 0–99%, 2–97% and 40–94% under anoxic, oxic, or anoxic/oxic treatments, respectively. The highest biodegradation of total PCB congeners occurred using Sybron under oxic and anoxic conditions (76.0% and 91.3% reduction, respectively; initial PCB concentration, 1417.1 mg l^{-1}). Also, the highest biodegradation extent of the PCB congeners occurred when the commercially available mixture of microorganisms, Sybron, was used (84.7% reduction) under combined anoxic/oxic conditions. Demonstration that biodegradation of most of the individual PCB congeners was achieved by the commercial mixtures of microorganisms in this study suggests that PCB-impacted environments

can sustain populations of these PCB-metabolizing organisms. This is particularly relevant for the development of biostimulation or bioaugmentation strategies for the bioremediation of PCB-contaminated wastes.

Keywords: Bioaugmentation; Bioremediation; Mixtures of microorganisms; PCBs; Transformer oil

Richard W. Eaton^{1*} and Peter Sandusky² . (Southern Regional Research Center, Agricultural Research Service, U.S. Department of Agriculture, New Orleans, Louisiana 70124,¹ Department of Environmental Science and Engineering, The University of North Carolina, Chapel Hill, North Carolina 27599² * Corresponding author. Mailing address: SRRC, ARS, USDA, 1100 Robert E. Lee Blvd., New Orleans, LA 70124. Phone: (504) 286-4353. Fax: (504) 286-4419. E-mail: richard.eaton@ars.usda.gov). **Biotransformations of 2-Methylisoborneol by Camphor-Degrading Bacteria** . *Applied and Environmental Microbiology*, Volume 75(3) (2009): 583-588

Many camphor-degrading bacteria that are able to transform 2-methylisoborneol (2-MIB) have been identified. Three of these strains have been examined in detail. *Rhodococcus ruber* T1 metabolizes camphor through 6-hydroxycamphor but converts 2-MIB to 3-hydroxy-2-MIB. *Pseudomonas putida* G1, which metabolizes camphor through 5-hydroxycamphor, converts MIB primarily to 6-hydroxy-2-MIB. *Rhodococcus wratislaviensis* DLC-cam converts 2-MIB through 5-hydroxy-2-MIB to 5-keto-2-MIB. Together, these three strains produce metabolites resulting from hydroxylation at all of the three available secondary carbons on the six-member ring of 2-MIB.

Iman A. El Gheriany,¹ Daniela Bocioaga,² Anthony G. Hay,² William C. Ghiorse,² Michael L. Shuler,¹ and Leonard W. Lion^{3*} (School of Chemical and Biomolecular Engineering,¹ Department of Microbiology,² School of Civil and Environmental Engineering, Cornell University, Ithaca, New York 14853³. * Corresponding author. Mailing address: School of Civil and Environmental Engineering, Hollister Hall, Cornell University, Ithaca, NY 14853. Phone: (607) 255-7571. Fax: (607) 255-9004. E-mail: LWL3@cornell.edu). **Iron Requirement for Mn(II) Oxidation by *Leptothrix discophora* SS-1** . *Applied and Environmental Microbiology*, Volume 75 (5) n(2009): 1229-1235

A common form of biocatalysis of Mn(II) oxidation results in the formation of biogenic Mn(III, IV) oxides and is a key reaction in the geochemical cycling of Mn. In this study, we grew the model Mn(II)-oxidizing bacterium *Leptothrix discophora* SS-1 in media with limited iron (0.1 μ M iron/5.8 mM pyruvate) and sufficient iron (0.2 μ M iron/5.8 mM pyruvate). The influence of iron on the rate of extracellular Mn(II) oxidation was evaluated. Cultures in which cell growth was limited by iron exhibited reduced abilities to oxidize Mn(II) compared to cultures in medium with sufficient iron. While the extracellular Mn(II)-oxidizing factor (MOF) is thought to be a putative multicopper oxidase, Mn(II) oxidation in the presence of zero added Cu(II) was detected and the decrease in the observed Mn(II) oxidation rate in iron-limited cultures was not relieved when the medium was supplemented with Cu(II). The decline of Mn(II) oxidation under iron-limited conditions was not accompanied by siderophore production and is unlikely to be an artifact of siderophore complex formation with Mn(III). The temporal variations in *mofA* gene transcript levels under conditions of limited and abundant iron were similar, indicating that iron

limitation did not interfere with the transcription of the *mofA* gene. Our quantitative PCR results provide a step forward in understanding the regulation of Mn(II) oxidation. The mechanistic role of iron in Mn(II) oxidation is uncertain; the data are consistent with a direct requirement for iron as a component of the MOF or an indirect effect of iron resulting from the limitation of one of many cellular functions requiring iron.

Susan Winch¹, Heath J. Mills², Joel E. Kostka³, Danielle Fortin¹ & David R.S. Lean⁴. (¹ Department of Earth Science, University of Ottawa, Ottawa, ON, Canada; ² Department of Oceanography, Texas A&M University, College Station, TX, USA; ³ Department of Oceanography, Florida State University, Tallahassee, FL, USA; and ⁴ Department of Biology, University of Ottawa, Ottawa, ON, Canada Correspondence: Susan Winch, Department of Earth Science, University of Ottawa, 985 Parkhurst Boulevard, Ottawa, ON, Canada K2A 3M8. . Tel.: +1 613 729 6751; fax: +1 613 721 0029; e-mail: swinch@magma.ca). **Identification of sulfate-reducing bacteria in methylmercury-contaminated mine tailings by analysis of SSU rRNA genes FEMS Microbiology Ecology, Volume 68(1) (2009): 94 – 107**

Sulfate-reducing bacteria (SRB) are often used in bioremediation of acid mine drainage because microbial sulfate reduction increases pH and produces sulfide that binds with metals. Mercury methylation has also been linked with sulfate reduction. Previous geochemical analysis indicated the occurrence of sulfate reduction in mine tailings, but no molecular characterization of the mine tailings-associated microbial community has determined which SRB are present. This study characterizes the bacterial communities of two geochemically contrasting, high-methylmercury mine tailing environments, with emphasis on SRB, by analyzing small subunit (SSU) rRNA genes present in the tailings sediments and in enrichment cultures inoculated with tailings. Novel *Deltaproteobacteria* and *Firmicutes*-related sequences were detected in both the pH-neutral gold mine tailings and the acidic high-sulfide base-metal tailings. At the subphylum level, the SRB communities differed between sites, suggesting that the community structure was dependent on local geochemistry. Clones obtained from the gold tailings and enrichment cultures were more similar to previously cultured isolates whereas clones from acidic tailings were more closely related to uncultured lineages identified from other acidic sediments worldwide. This study provides new insights into the novelty and diversity of bacteria colonizing mine tailings, and identifies specific organisms that warrant further investigation with regard to their roles in mercury methylation and sulfur cycling in these environments.

Keywords: sulfate-reducing bacteria • mine tailings • mercury • SSU rRNA • microbial community

Xuesong Zhao^{1, 2}, Juan Wang¹, Jie Li¹, Ling Fu¹, Juan Gao¹, Xiuli Du¹, Hongtao Bi¹, Yifa Zhou¹ and Guihua Tai¹. (¹Laboratory of Molecular Epigenetics of MOE, School of Life Sciences, Northeast Normal University, 130024 Changchun, People's Republic of China, ²Fuxin Advanced Polytechnic College, 123000 Fuxin, People's Republic of China. **Yifa Zhou (Corresponding author) Email: zhouyf383@nenu.edu.cn). Highly selective biotransformation of ginsenoside Rb1 to Rd by the phytopathogenic fungus *Cladosporium fulvum* (syn. *Fulvia fulva*). *Journal of Industrial Microbiology and Biotechnology*, Volume 36(5) (2009): 721-726**

Fourteen phytopathogenic fungi were tested for their ability to transform the major ginsenosides to the active minor ginsenoside Rd. The transformation products were identified by TLC and

HPLC, and their structures were assigned by NMR analysis. *Cladosporium fulvum*, a tomato pathogen, was found to transform major ginsenoside Rb₁ to Rd as the sole product. The following optimum conditions for transforming Rd by *C. fulvum* were determined: the time of substrate addition, 24 h; substrate concentration, 0.25 mg ml⁻¹; temperature, 37°C; pH 5.0; and biotransformation period, 8 days. At these optimum conditions, the maximum yield was 86% (molar ratio). Further, a preparative scale transformation with *C. fulvum* was performed at a dose of 100 mg of Rb₁ by a yield of 80%. This fungus has potential to be applied on the preparation for Rd in pharmaceutical industry.

Keywords: Biotransformation - *Cladosporium fulvum* (syn. *Fulvia fulva*) - Ginsenoside Rb₁ - Ginsenoside Rd

Yanliang Lin, Xin Song^{*}, Juan Fu, Jianqiang Lin, Yinbo Qu. (State Key Laboratory of Microbial Technology, Shandong University, Jinan 250100 China. ^{*}Correspondence to Xin Song, State Key Laboratory of Microbial Technology, Shandong University, Jinan 250100 China. email: Xin Song (songx@sdu.edu.cn)). Microbial transformation of androst-4-ene-3, 17-dione by *Bordetella* sp. B4 CGMCC 2229 *Journal of Chemical Technology & Biotechnology*, Volume 84(5) (2009): 789 – 793

BACKGROUND: Microbial transformation of steroids has attracted widespread attention, especially the transformation of those steroids synthesized with difficulty by chemical methods. In this study, microbial transformation of androst-4-ene-3, 17-dione (AD) by *Bordetella* sp. B4 was investigated, and the effect of temperature on transformation was studied.

RESULTS: Three metabolites were purified by preparative TLC and HPLC, and identified as androsta-1,4-diene-3,17-dione (ADD), 9 α -hydroxyandrost-4-ene-3, 17-dione (9 α -OH-AD), and 3-hydroxy-9, 10-secoandrost-1, 3, 5-triene-9, 17-dione (3-OH-SATD) by nuclear magnetic resonance imaging (NMR), Fourier transform infrared spectroscopy (FTIR) and mass spectroscopy (MS). It was first reported that the genus of *Bordetella* has the capability of AD degradation. Microbial transformation of AD was performed at 30 °C, 37 °C, 40 °C and 45 °C. The 9 α -OH-AD yield reached a maximum within 16 h when the strain was cultivated in media with AD as sole carbon at 37 °C. Surprisingly, ADD was produced by the strain cultivated at 40 °C but not at 37 °C, which was different from previous reports. It was deduced that the alcohol dehydrogenase that catalyzed the transformation of AD to ADD may be temperature sensitive.

CONCLUSION: Androst-4-ene-3,17-dione was converted into 9 α -hydroxyandrost-4-ene-3, 17-dione and other metabolites rapidly by *Bordetella* sp. B4. It is anticipated that the strain *Bordetella* sp. B4 CGMCC 2229 can be used in the steroids industry.

Keywords: *Bordetella* sp. B4 CGMCC 2229 • androst-4-ene-3,17-dione • 9 α -hydroxyandrost-4-ene-3,17-dione • androsta-1,4-diene-3,17-dione • biotransformation

Adrian Hernandez-Mendoza^{1 2}, Arnoldo Lopez-Hernandez^{1 2}, Charles G Hill², Hugo S Garcia^{1 2 *}. (¹UNIDA-Instituto Tecnológico de Veracruz, MA de Quevedo #2779, Col. Formando Hogar, Veracruz, Ver. 91897, Mexico, ²Department of Chemical and Biological Engineering, University of Wisconsin, 1415 Engineering Drive, Madison, WI 53706, USA. email: Hugo S Garcia (hsgarcia@itver.edu.mx). Bioconversion of linoleic acid to

conjugated linoleic acid by *Lactobacillus reuteri* under different growth conditions. Journal of Chemical Technology & Biotechnology, Volume 84(2) (2009): 180 – 185

BACKGROUND: *Lactobacillus reuteri* was grown in De Man/Rogosa/Sharpe (MRS) broth (initial pH 6.5) supplemented with free linoleic acid (LA) at different concentrations (5, 10, 20 and 30 mg mL⁻¹) and incubated aerobically at different temperatures (4, 10, 16, 22 and 30 °C) in order to test its ability to accomplish the bioconversion of LA to conjugated linoleic acid (CLA). Temperatures and LA concentrations producing the highest conversion of LA to CLA in the initial trials were tested further using micro-anaerobic conditions and a lower initial pH (5.5).

RESULTS: Data showed that production of CLA exhibited variations with regard to the fermentation conditions used. The highest production of CLA (0.108 mg mL⁻¹) was measured in a broth containing 20 mg mL⁻¹ free LA that was incubated aerobically at 10 °C for 30 h. When the initial pH of the reaction medium was reduced from 6.5 to 5.5, CLA production decreased. Micro-aerobic conditions reduced the ability of *Lb. reuteri* to produce CLA, since production of CLA under aerobic conditions was at least 1.4 times greater.

CONCLUSION: Production of CLA by *Lb. reuteri* at low temperatures and relatively high substrate concentrations provides novel opportunities for the development of functional foods with the benefits of enrichment in CLA and probiotic bacteria.

Keywords: *Lactobacillus reuteri* • conjugated linoleic acid • bioconversion • probiotic • linoleic acid

Jiu-Hong Li, Yi-Xin Guan^{*}, Hai-Qing Wang, Shan-Jing Yao. (*Correspondence to Yi-Xin Guan, Department of Chemical and Biochemical Engineering, Zhejiang University, Hangzhou 310027, China). Dehydrogenation of 11 α -hydroxy-16 α , 17-epoxyprogesterone by encapsulated *Arthrobacter simplex* cells in an aqueous/organic solvent two-liquid-phase system. Journal of Chemical Technology & Biotechnology, Volume 84(2) (2009): 208 - 214

BACKGROUND: *Arthrobacter simplex* cells immobilised in sodium cellulose sulfate/polydimethyl-diallyl-ammonium chloride microcapsules were used for the microbial dehydrogenation of 11 α -hydroxy-16 α ,17-epoxyprogesterone to 11 α -hydroxy-16 α ,17-epoxypregn-1,4-diene-3,20-dione in an aqueous/organic solvent two-liquid-phase system, which is a key reaction in the production of glucocorticoid pharmaceuticals. The aim of the study was to establish a suitable aqueous/organic solvent two-liquid-phase system for performing semi-continuous production in an airlift loop reactor by encapsulated *A. simplex* cells with the addition of suitable surfactants to achieve a higher yield of the product.

RESULTS: *n*-Hexane was selected as the most suitable organic solvent. In optimised Tween-80 emulsion feed mode the conversion in the airlift loop reactor was as high as 97.54% when the time of reaction was 2 h, and the reaction time was greatly shortened. In semi-continuous production the cultivation with immobilised cells was carried out for five batches in total. The conversion in each batch was above 95% and the enzymatic activity still remained quite high after five batches of biotransformation

CONCLUSION: The results showed that performing the conversion by this method shortened the reaction time and increased the productivity, thus demonstrating the great potential of the method for the dehydrogenation of 11 α -hydroxy-16 α ,17-epoxyprogesterone.

Keywords: *Arthrobacter simplex* • Tween-80 emulsion feed mode • sodium cellulose sulfate (NaCS)/poly-dimethyl-diallyl-ammonium chloride (PDMDAAC) microcapsule • aqueous/organic solvent two-liquid-phase system • semi-continuous biotransformation

Marthah De Lorme¹ and Morrie Craig². (¹Department of Microbiology, Oregon State University, 1007 Agricultural and Life Sciences Building, Corvallis, OR 97331-4501, USA, ²College of Veterinary Medicine, Oregon State University, Corvallis, OR 97331, USA). **Biotransformation of 2,4,6-Trinitrotoluene by Pure Culture Ruminant Bacteria. Current Microbiology, Volume 58(1) (2009): 81-86**

Twenty-one ruminal bacteria species were tested for their ability to degrade 2,4,6-trinitrotoluene (TNT) within 24 h. *Butyrivibrio fibrisolvens*, *Fibrobacter succinogenes*, *Lactobacillus vitulinus*, *Selenomonas ruminantium*, *Streptococcus caprinus*, and *Succinivibrio dextrinosolvens* were able to completely degrade 100 mg/L TNT, with <5% of the original TNT recovered as diaminonitrotoluene metabolites. *Eubacterium ruminantium*, *Lactobacillus ruminis*, *Ruminobacter amylophilus*, *Streptococcus bovis*, and *Wolinella succinogenes* were able to completely degrade 100 mg/L TNT, with 23–60% of the TNT recovered as aminodinitrotoluene and/or diaminonitrotoluene metabolites. *Clostridium polysaccharolyticum*, *Megasphaera elsdenii*, *Prevotella bryantii*, *Prevotella ruminicola*, *Ruminococcus albus*, and *Ruminococcus flavefaciens* were able to degrade 80–90% of 100 mg/L TNT. *Desulfovibrio desulfuricans* subsp. *desulfuricans*, *Prevotella albensis*, and *Treponema bryantii* degraded 50–80% of the TNT. *Anaerovibrio lipolytica* was completely inhibited by 100 mg/L TNT. These results indicate that a variety of rumen bacteria is capable of transforming TNT.

Hiroshi Habe¹, Tokuma Fukuoka¹, Dai Kitamoto¹ and Keiji Sakaki¹. (¹Research Institute for Innovations in Sustainable Chemistry, National Institute of Advanced Industrial Science and Technology (AIST), Tsukuba Central 5-2, 1-1-1 Higashi, Tsukuba Ibaraki, 305-8565, Japan). **Biotransformation of glycerol to d -glyceric acid by *Acetobacter tropicalis*. Applied Microbiology and Biotechnology, Volume 81(6) (2009): 1033-1039**

Bacterial strains capable of converting glycerol to glyceric acid (GA) were screened among the genera *Acetobacter* and *Gluconacetobacter*. Most of the tested *Acetobacter* and *Gluconacetobacter* strains could produce 1.8 to 9.3 g/l GA from 10% (v/v) glycerol when intact cells were used as the enzyme source. *Acetobacter tropicalis* NBRC16470 was the best GA producer and was therefore further investigated. Based on the results of high-performance liquid chromatography analysis and specific rotation, the enantiomeric composition of the produced GA was D-glyceric acid (D-GA). The productivity of D-GA was enhanced with the addition of both 15% (v/v) glycerol and 20 g/l yeast extract. Under these optimized conditions, *A. tropicalis* NBRC16470 produced 22.7 g/l D-GA from 200 g/l glycerol during 4 days of incubation in a jar fermentor.

Keywords: Glycerol use - D-glyceric acid - Acetic acid bacteria - *Acetobacter* sp. - *Gluconacetobacter* sp.

Jen-Chieh Tsai^a, Mathava Kumar^a and Jih-Gaw Lin^a. (^aInstitute of Environmental Engineering, National Chiao Tung University, 75 Po-Ai Street, Hsinchu 300, Taiwan, ROC). **Anaerobic biotransformation of fluorene and phenanthrene by sulfate-reducing**

bacteria and identification of biotransformation pathway. Journal of Hazardous Materials, Volume 164(2-3) (2009): 847-855

In the present study, anaerobic biotransformation of fluorene and phenanthrene by sulfate-reducing bacteria (SRB) was investigated and biotransformation pathways were proposed. SRB was enriched from anaerobic swine wastewater sludge and its abundance was determined by the fluorescence in situ hybridization (FISH) technique. Batch anaerobic biotransformation studies were conducted with fluorene (5 mg L⁻¹), phenanthrene (5 mg L⁻¹) and a mixture of the two (10 mg L⁻¹). After 21 d of incubation, 88% of fluorene and 65% of phenanthrene were biotransformed by SRB. In contrast to previous studies, a decrease in biotransformation efficiency was observed in the presence of both fluorene and phenanthrene. Throughout the study, sulfate reduction was coupled with biotransformation of fluorene and phenanthrene. However, no increase in bacterial cell density was observed in the presence of an inhibitor, i.e. molybdate. Identification of metabolites by gas chromatography–mass spectrometry (GC–MS) revealed that fluorene and phenanthrene were biotransformed through a sequence of hydration and hydrolysis reactions followed by decarboxylation with the formation of *p*-cresol (only in the phenanthrene system) and phenol. The metabolites identified suggest novel biotransformation pathways of fluorene and phenanthrene.

Keywords: Anaerobic biotransformation; Fluorene; Phenanthrene; Sulfate-reducing bacteria; Biotransformation pathway

Biomarker

M. Ashraf^a. (^aDepartment of Botany, University of Agriculture, Faisalabad, Pakistan). Biotechnological approach of improving plant salt tolerance using antioxidants as markers. Biotechnology Advances, Volume 27(1) (2009): 84-93

Salt stress causes multifarious adverse effects in plants. Of them, production of reactive oxygen species (ROS) is a common phenomenon. These ROS are highly reactive because they can interact with a number of cellular molecules and metabolites thereby leading to a number of destructive processes causing cellular damage. Plants possess to a variable extent antioxidant metabolites, enzymes and non-enzymes, that have the ability to detoxify ROS. In the present review, the emphasis of discussion has been on understanding the role of different antioxidants in plants defense against oxidative stress caused by salt stress. The role of different antioxidants as potential selection criteria for improving plant salt tolerance has been critically discussed. With the advances in molecular biology and availability of advanced genetic tools considerable progress has been made in the past two decades in improving salt-induced oxidative stress tolerance in plants by developing transgenic lines with altered levels of antioxidants of different crops. The potential of this approach in counteracting stress-induced oxidative stress has been discussed at length in this review.

Keywords: Salt stress; Reactive oxygen species; Enzymatic and non-enzymatic antioxidants

Barbara Kasprzyk-Hordern^{a, b}, Richard M. Dinsdale^b and Alan J. Guwy^b. (^aUniversity of Huddersfield, Department of Chemical and Biological Sciences, Queensgate, Huddersfield HD1 3DH, UK, ^bUniversity of Glamorgan, Sustainable Environment Research Centre, Faculty of Health, Sport and Science, Pontypridd CF37 1DL, UK). Illicit drugs and

pharmaceuticals in the environment – Forensic applications of environmental data, Part 2: Pharmaceuticals as chemical markers of faecal water contamination. Environmental Pollution, Volume 157(60) (2009) : 1778-1786

This manuscript is part two of a two-part study aiming to provide a better understanding and application of environmental data not only for environmental aims but also to meet forensic objectives. In this paper pharmaceuticals were investigated as potential chemical indicators of water contamination with sewage. The monitoring program carried out in Wales revealed that some pharmaceuticals are particularly persistent and/or ubiquitous in contaminated river water and therefore might be considered as potential conservative or labile wastewater indicators. In particular, these include some anti-inflammatory/analgesics, antiepileptics, beta-blockers, some H2-receptor antagonists and antibacterial drugs.

Wastewater as an indicative source of information can be used in forensic applications.

Keywords: Pharmaceuticals; Surface water; Wastewater markers; Chemical tracers; Environmental forensics

Rebecca Klaper^a, Jordan Crago^a, Jessica Barr^a, Devrah Arndt^a, Kristina Setyowati^b and Jian Chen^b. (^aGreat Lakes WATER Institute, University of Wisconsin-Milwaukee, 600 East Greenfield Ave., Milwaukee, WI 53204, USA, ^bDepartment of Chemistry, University of Wisconsin-Milwaukee, 3210 N. Cramer Street, Milwaukee, WI 53201, USA). **Toxicity biomarker expression in daphnids exposed to manufactured nanoparticles: Changes in toxicity with functionalization. Environmental Pollution, Volume 157(4) (2009) : 1152-1156**

In previous work we have shown that the toxicity of nanomaterials to *Daphnia* spp. differs with the type of nanoparticle either due to the core of the particle or to the way in which a particle suspension is prepared. The purpose of this study was to investigate the toxicity and antioxidant response of *Daphnia pulex* in relation to a change in surface functionalization of nanomaterials with the same core material, nC60. Despite the lack of acute toxicity for various nC60 suspensions up to 100 ppm concentration, there was a significant production of the toxicity biomarkers glutathione-S-transferase and catalase, at lower concentrations indicating changes in reactive oxygen species. Nanoparticle functionalization significantly affected this response. Oxidative stress markers appear to be a good predictor of potential future toxicity of nanomaterials. Functionalization alters both toxicity and oxidative stress in whole organism assays.

Antioxidant response of *Daphnia* to nanoparticles with differing surface functionalization and core structure.

Keywords: *Daphnia pulex*; Toxicity; Nanoparticle; Glutathione-S-transferase; Oxidative stress

Giuseppe Ungherese^a and Alberto Ugolini^a. (^aDipartimento di Biologia Evoluzionistica, Università di Firenze, Via Romana 17, 50125 Firenze, Italy). **Sandhopper solar orientation as a behavioural biomarker of trace metals contamination. Environmental Pollution, Volume 157(4) (2009) : 1360-1364**

Although many studies have focused on trace metals accumulation, investigations of talitrid amphipods as biomarkers are rare. This study explores the possibility of using the solar orientation capacity of *Talitrus saltator* as a behavioural marker of exposure to two essential (Cu and Zn) and two non-essential (Cd and Hg) metals. LC₅₀ analyses performed before the solar orientation tests showed that the 72 h LC₅₀ for Hg was 0.02 ppm while the 96 h LC₅₀ values for Cu, Cd and Zn were 13.28 ppm, 27.66 ppm, and 62.74 ppm, respectively. The presence of metals in seawater affects the solar orientation capacity of *T. saltator* in a concentration-dependent manner and according to the toxicity ranking of the metals (Hg > Cu > Cd > Zn). Therefore, the solar orientation capacity of *T. saltator* seems to be a promising behavioural marker for exposure to trace metals.

Solar orientation capacity is a promising behavioural marker for exposure to trace metals in sandhoppers.

Keywords: Amphipods; *Talitrus saltator*; Trace metals; Biomarker; Behaviour

Biofertilizer

Muhammad Yasir^a, Zubair Aslam^a, Seon Won Kim^a, Seon-Woo Lee^b, Che Ok Jeon^c and Young Ryun Chung^a. (^aDivision of Applied Life Science (BK 21), PMBBRC and EB-NCRC, Gyeongsang National University, Jinju 660-701, Republic of Korea, ^bDepartment of Applied Biology, College of Natural Resources and Life Science, Dong-A University, Busan 604-714, Republic of Korea, ^cDepartment of Life Sciences, Chung-Ang University, Seoul 156-756, Republic of Korea). **Bacterial community composition and chitinase gene diversity of vermicompost with antifungal activity. Bioresource Technology, Volume 100(19) (2009): 4396-4403**

Bacterial communities and chitinase gene diversity of vermicompost (VC) were investigated to clarify the influence of earthworms on the inhibition of plant pathogenic fungi in VC. The spore germination of *Fusarium moniliforme* was reduced in VC aqueous extracts prepared from paper sludge and dairy sludge (fresh sludge, FS). The bacterial communities were examined by culture-dependent and -independent analyses. Unique clones selected from 16S rRNA libraries of FS and VC on the basis of restriction fragment length polymorphism (RFLP) fell into the major lineages of the domain bacteria *Proteobacteria*, *Bacteroidetes*, *Verrucomicrobia*, *Actinobacteria* and *Firmicutes*. Among culture isolates, *Actinobacteria* dominated in VC, while almost equal numbers of *Actinobacteria* and *Proteobacteria* were present in FS. Analysis of chitinolytic isolates and chitinase gene diversity revealed that chitinolytic bacterial communities were enriched in VC. Populations of bacteria that inhibited plant fungal pathogens were higher in VC than in FS and particularly chitinolytic isolates were most active against the target fungi.

Keywords: Bacterial community; Chitinase; Plant pathogenic fungi; Paper sludge; Vermicomposting

Cheng-Hsiung Chang^a and Shang-Shyng Yang^{a, b}. (^aInstitute of Microbiology and Biochemistry, National Taiwan University, Taipei 10617, Taiwan, ^bDepartment of Biochemical Science and Technology, National Taiwan University, Taipei 10617, Taiwan). **Thermo-tolerant phosphate-solubilizing microbes for multi-functional biofertilizer preparation. Bioresource Technology, Volume 100(4) (2009): 1648-1658**

In order to prepare the multi-functional biofertilizer, thermo-tolerant phosphate-solubilizing microbes including bacteria, actinomycetes, and fungi were isolated from different compost plants and biofertilizers. Except *Streptomyces thermophilus* J57 which lacked pectinase, all isolates possessed amylase, CMCase, chitinase, pectinase, protease, lipase, and nitrogenase activities. All isolates could solubilize calcium phosphate and Israel rock phosphate; various isolates could solubilize aluminum phosphate, iron phosphate, and hydroxyapatite. During composting, biofertilizers inoculated with the tested microbes had a significantly higher temperature, ash content, pH, total nitrogen, soluble phosphorus content, and germination rate than non-inoculated biofertilizer; total organic carbon and carbon-to-nitrogen ratio showed the opposite pattern. Adding these microbes can shorten the period of maturity, improve the quality, increase the soluble phosphorus content, and enhance the populations of phosphate-solubilizing and proteolytic microbes in biofertilizers. Therefore, inoculating thermo-tolerant phosphate-solubilizing microbes into agricultural and animal wastes represents a practical strategy for preparing multi-functional biofertilizer.

Keywords: Thermo-tolerant phosphate-solubilizing microbes; Soluble phosphorus content; Biofertilizer; Maturity

María del Carmen Rivera-Cruz^a, Antonio Trujillo Narcía^b, Georgina Córdova Ballona^a, Josef Kohler^c, Fuensanta Caravaca^c and Antonio Roldán^c. (^aColegio de Postgraduados, Department of Environmental Microbiology, Campus de Tabasco, 86500 Cárdenas, Mexico, ^bUniversidad Autónoma de Guadalajara, Campus de Tabasco, 86039 Villahermosa, Mexico, ^cCSIC-Centro de Edafología y Biología Aplicada del Segura, Department of Soil and Water Conservation, P.O. Box 164, Campus de Espinardo, 30100 Murcia, Spain). **Poultry manure and banana waste are effective biofertilizer carriers for promoting plant growth and soil sustainability in banana crops. *Soil Biology and Biochemistry*, Volume 40(12) (2008): 3092-3095**

The aims of our study were to compare the effectiveness of poultry manure (PM) and banana waste (BW), with regard to their use as inoculant carriers of a bacterial consortium constituted by strains of *Azospirillum*, *Azotobacter* and P-solubiliser bacteria and to establish the most efficient dose of biofertilizer for a soil cultivated with banana (*Musa paradisiaca* AAA Simmonds), with respect to improving plant performance and soil physical and microbiological properties. Six months after planting, plant growth had increased with increase in dose of the biofertilizers applied. The biofertilizer prepared on BW enhanced the density of P-solubiliser bacteria, the concentrations of available P and foliar P to a greater extent than with the biofertilizer prepared on PM. The increases produced by the biofertilizer prepared on PM for the soil aggregate stability, enzymatic activities and the labile carbon fractions were highly correlated to the dose applied. Both biofertilizers can be considered potentially useful as inoculant carriers of PGPR but the usefulness of BW appears to be restricted to moderate doses of application ($\leq 3\%$).

Keywords: Aggregate stability; *Azospirillum*; *Azotobacter*; Biofertilizer; Enzymatic activity; PGPR

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Av. San Martín 4457, Buenos Aires, Argentina, ^bDZD Agro SRL, Av. San Martín 430, (6237) América, Buenos Aires, Argentina). Field performance of a liquid formulation of *Azospirillum brasilense* on dryland wheat productivity. *European Journal of Soil Biology*, Volume 45(1) (2009): 3-11

The beneficial effects of inoculating with *Azospirillum brasilense* on crop productivity have been widely described, but extensive use in typical agricultural field environments is scarcely documented. The objective of this study was to quantify the productivity of wheat (*Triticum aestivum* L.) whose seed was inoculated with a liquid formulation containing *Azospirillum brasilense* INTA Az-39 strain under typical dryland farming conditions. The study was performed in the 2002–2006 growing seasons, evaluating inoculated and non-inoculated seed at 297 experimental locations in the Pampas region of Argentina. The inoculated crops exhibited more vigorous vegetative growth, with both greater shoot and root dry matter accumulation (12.9 and 22.0%, respectively). The inoculation increased the number of harvested grains by 6.1%, and grain yield by 260 kg ha⁻¹ (8.0%). Positive responses were determined in about 70% of the sites, depending mostly on the attainable yield and independently of fertilization and other crop and soil management practices. In general, more response to inoculation was observed in the absence of major crop growth limitations, suggesting the complementary contribution of the *Azospirillum brasilense* treatment to more efficiently developing higher yielding wheat.

Keywords: Biofertilizer; Subhumid and Semiarid regions; Mollisols; Grain yield components

Anton Hartmann^a and Yoav Bashan^b. (^aHelmholtz Zentrum München, German Research Centre for Environmental Health, Department Microbe-Plant Interactions, Ingolstaedter Landstr. 1, D-85764 Neuherberg, Germany, ^bCIBNOR, Environmental Microbiology Group, Mar Bermejo 195, Col. Playa Palo de Santa Rita, La Paz, BCS 23090, Mexico Fax: +52 612 1254710). Ecology and application of *Azospirillum* and other plant growth-promoting bacteria (PGPB) – Special Issue. *European Journal of Soil Biology*, Volume 45(1) (2009): 1-2

Plant growth-promoting bacteria of non-leguminous plants have been the focus of research since several decades because non-leguminous plants like rice, wheat, maize – to name just the most prominent ones – are the most important crops feeding the ever growing human population on our planet. Leguminous plants harbour specific symbiotic systems with root or stem-nodulating rhizobia for plant growth promotion and nitrogen fixation, which are characterized by high host specificity of plant-bacteria interaction, such that it was not possible until now to widen the application range of rhizobia beyond this evolutionary boundary.

The major motif to contribute to sustainable support of non-leguminous crop plant growth with less fertilizer and agrochemical input led Dr. Johanna Döbereiner and her research group at the EMBRAPA-Institute of Agrobiologia in Seropedica, Rio de Janeiro, Brasil, to investigate root-associated nitrogen fixing bacteria of non-leguminous crop plants in the 1960 and 1970s. This resulted in the rediscovery of the rhizosphere diazotrophic bacterial species *Azospirillum brasilense* and *Azospirillum lipoferum*, published in 1978. In the following years and decades, many more very interesting root surface colonizing and endophytic diazotrophic PGPB were discovered and characterized from this and other laboratories. The search for prominent and potent diazotrophic PGPB and their diversity in nature is still a fascinating ongoing research field.

Physiological and biochemical research of basic metabolic features of *A. brasilense* and its plant growth promotion potential was pioneered also in the late 1970s by Dr. Yaacov Okon when he was pursuing post-doc in Prof. R.H. Burris laboratory at the University of Madison, Wisconsin, USA and continued at the Hebrew University of Jerusalem, Faculty of Agriculture in Rehovot, Israel. Important biochemical and molecular genetic research on *Azospirillum* and its interaction with plants were contributed in the 1980s also by several other labs working on *A. brasilense* and other diazotrophic PGPB. Currently, much expectation is on the almost completed genomic sequencing projects of *A. brasilense* Sp245 in U.S.A. and *A. lipoferum* 4B in France. The genomes of other diazotrophic PGPB, like *Azoarcus* sp. BH52, *Herbaspirillum seropedicae* and *Gluconacetobacter diazotrophicus* were recently completed enabling new insights and starting points of even more advanced studies.

On the other hand, *Azospirillum* has been the focus of applied research also from the 1970s of last century. It became soon apparent, that plant growth promotion has also much to do with phytohormonal and phytosanitary mechanisms of microbial interactions in the rhizosphere and microbe-plant interactions with the potential for substantial stimulation of plant development and health. The practical application as inoculant (biofertilizer) was a quite controversial one from the beginning, because the growth response of crops is not completely predictable in its extent and depends – not surprisingly – on many ecological and agrobiotechnological factors. Nevertheless, much progress has been made also in this field, leading to an ever growing and successful application of *Azospirillum* and PGPB in several regions of the world, especially in South and Central America. The leading countries in practical field applications of *Azospirillum* are Mexico with estimated 300,000 ha inoculated fields in 2007, and Argentina where over 220,000 ha of wheat and corn were commercially inoculated with *Azospirillum* in 2008. This development most recently coincided with the dramatic raise in the oil price and costs of chemical nitrogen fertilizer as well as the fast growing importance of plant based “biofuels” in the context of the sought for solutions of the global climate change crisis.

A new emerging field for these bacteria is their potential use in solving environment problems such as assisting reforestation efforts of severely eroded lands, restoration of marine mangrove ecosystems, phytostabilization of mine tailings, assisting in metal and pesticide decontamination of soils and biological treatment of wastewater where they enhance the capacity of microalgae in recycling of wastewater.

Already in the early 1980s, a series of scientific workshops on “*Azospirillum*: genetics, physiology, ecology” have been initiated and organized in Germany at the Institute of Genetics, University of Bayreuth by Prof. Dr. Walter Klingmüller and coworkers, continued by workshops in Hannover, Germany and Sárvár, Hungary, by Dr. Istvan Fendrik, University of Hannover (Germany). This series was lately continued in September 2007 by the “VIIth *Azospirillum* and related PGPR”-workshop organized by Prof. Yvan Moenne-Loccoz (University of Lyon, France) and Prof. Bruno Touraine (University of Montpellier, France) and their teams as satellite meeting of the “Rhizosphere II” symposium in Montpellier, France. In October 2007, there was another prominent workshop on *Azospirillum* in Argentina “First International workshop on *Azospirillum*” that was organized by Dr. Fabricio Cassan, Dr. Claudio Penna and the network for quality control of inoculants of the Argentinean Association of Microbiology in Cordoba, Argentina.

This special issue contains contributions to these two workshops combined with other contributions in this research field. We thank the editor in chief, Prof. Dr. Christoph Tebbe and the editorial staff, especially Michelle Paczy, for their great support and dedication in preparing this very timely special issue on “Ecology and application of *Azospirillum* and other PGPB”.

Biocomposting

D.K. Sharma^b, A.K. Pandey^a and Lata^a. (^aDivision of Microbiology, Indian Agricultural Research Institute, New Delhi 110012, India, ^bDivision of Environmental Sciences, Indian Agricultural Research Institute, New Delhi 110012, India). **Use of *Jatropha curcas* hull biomass for bioactive compost production. *Biomass and Bioenergy*, Volume 33(1) (2009): 159-162**

The paper deals with utilization of biomass of *Jatropha* hulls for production of bioactive compost. In the process of *Jatropha* oil extraction, a large amount of hull waste is generated. It has been found that the direct incorporation of hull into soil is considerably inefficient in providing value addition to soil due to its unfavorable physicochemical characteristics (high pH, EC and phenolic content). An alternative to this problem is the bioconversion of *Jatropha* hulls using effective lignocellulolytic fungal consortium, which can reduce the phytotoxicity of the degraded material. Inoculation with the fungal consortium resulted in better compost of *Jatropha* hulls within 1 month, but it takes nearly 4 months for complete compost maturation as evident from the results of phytotoxicity test. Such compost can be applied to the acidic soil as a remedial organic manure to help maintaining sustainability of the agro-ecosystem. Likewise, high levels of cellulolytic enzymes observed during bioconversion indicate possible use of fungi for ethanol production from fermentation of hulls.

Keywords: *Jatropha* hulls; Composting; Lignocellulolytic fungi; Phytotoxicity

Surindra Suthar^a. (^aEnvironmental Biology Laboratory, Post Graduate Department of Zoology, B.R.G. Govt. Girls College, Sri Ganganagar 335001, India). **Vermicomposting of vegetable-market solid waste using *Eisenia fetida*: Impact of bulking material on earthworm growth and decomposition rate *Ecological Engineering*, Volume 35(5) (2009) : 914-920**

Vegetable-market solid waste is produced in millions of tones in urban areas and creates a problem of safe disposal. The aim of this study was to convert vegetable solid waste (VW) amended with wheat straw (WS), cow dung (CD), and biogas slurry (BGS) into vermicompost using earthworm *Eisenia fetida*. VW was mixed in bulky materials (WS, CD, and BGS) in different ratios to produce eight different combinations for laboratory screening of wastes for 15 weeks. The vermicomposting caused a decrease in organic C (12.7–28%) and C:N ratio (42.4–57.8%), while increase in total N (50.6–75.8%), available P (42.5–110.4%), and exchangeable K (36.0–78.4%) contents. Waste mineralization and humification rates were higher in bedding those containing easy digestible bulky agents, i.e., BGS and CD. Worm-processed material obtained from BGS:VW (1:2) vermibed showed the higher total N (31.3 g kg⁻¹), available P (8.7 g kg⁻¹) and exchangeable K (20.7 g kg⁻¹) contents. The nutrient-rich vermicompost with acceptable C:N ratio ranges ($\geq 1:20$) indicates its agronomic potentials. Waste mixtures also supported the earthworm growth and reproduction rates in vermibeds. The results indicated that vermicomposting can be an efficient technology to convert negligible vegetable-market solid wastes into nutrient-rich biofertilizer if mixed with bulking materials in appropriate ratios.

Keywords: Vermicompost; Crop residues; Cow dung; Vegetable solid waste; *Eisenia fetida*; Cocoon; Urban solid wastes

Biopesticides

Véronique Edel-Hermann¹, Sylvie Brenot¹, Nadine Gautheron¹, Sébastien Aimé¹, Claude Alabouvette¹ & Christian Steinberg¹. (¹ INRA, Université de Bourgogne, UMR1229 Microbiologie du Sol et de l'Environnement, CMSE, Dijon, France. **Correspondence: Véronique Edel-Hermann, INRA, Université de Bourgogne, UMR1229 Microbiologie du Sol et de l'Environnement, CMSE, 17 rue Sully, BP 86510, F-21065 Dijon, France. Tel.: +33 3 80 69 34 50; fax: +33 3 80 69 32 24; e-mail: veronique.edel@dijon.inra.fr Editor: Christoph Tebbe).** **Ecological fitness of the biocontrol agent *Fusarium oxysporum* Fo47 in soil and its impact on the soil microbial communities. FEMS Microbiology Ecology, Volume 68(1) (2009): 37 – 45**

Some nonpathogenic strains of *Fusarium oxysporum* can control *Fusarium* diseases responsible for severe damages in many crops. Success of biological control provided by protective strains requires their establishment in the soil. The strain Fo47 has proved its efficacy under experimental conditions, but its ecological fitness has not been carefully studied. In a series of microcosm studies, the ability of a benomyl-resistant mutant Fo47b10 to establish in two different soils was demonstrated. One year after its introduction at two concentrations in the disinfected soils, the biocontrol agent (BCA) established at similar high population densities, whereas in the nondisinfected soils it survived at lower densities, related to the initial concentrations at which it was introduced. The BCA behaved similarly in the two soils at temperatures ranging from 5 to 25 °C and soil water potentials between –0.01 and –1.5 MPa. In addition, terminal restriction fragment length polymorphism analysis of 16S and 18S rRNA showed that the structures of the bacterial and fungal communities evolved with time but were not significantly affected by the introduction of the BCA. Overall, the results showed that Fo47 is potentially a good BCA, able to establish in different soil environments without perturbing the investigated microbial structures.

Keywords: biological control • ecological fitness • *Fusarium oxysporum* Fo47 • microbial community • risk assessment • terminal restriction fragment length polymorphism (T-RFLP)

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Microbial modification of polyunsaturated fatty acids can often lead to special changes in their structure and in biological potential. Therefore, the aim of this study was to develop potential

antifungal agents through the microbial conversion of docosahexaenoic acid (DHA). Bioconverted oil extract of docosahexaenoic acid (bDHA), obtained from the microbial conversion of docosahexaenoic acid (DHA) by *Pseudomonas aeruginosa* PR3, was assessed for its in vitro and in vivo antifungal potential. Mycelial growth inhibition of test plant pathogens, such as *Botrytis cinerea*, *Colletotrichum capsici*, *Fusarium oxysporum*, *Fusarium solani*, *Phytophthora capsici*, *Rhizoctonia solani* and *Sclerotinia sclerotiorum*, was measured in vitro. bDHA (5 µl disc⁻¹) inhibited 55.30–65.90% fungal mycelium radial growth of all the tested plant pathogens. Minimum inhibitory concentrations (MICs) of bDHA against the tested plant pathogens were found in the range of 125–500 µg ml⁻¹. Also, bDHA had a strong detrimental effect on spore germination for all the tested plant pathogens. Further, three plant pathogenic fungi, namely *C. capsici*, *F. oxysporum* and *P. capsici*, were subjected to an in vivo antifungal screening. bDHA at higher concentrations revealed a promising antifungal effect in vivo as compared to the positive control oligochitosan. Furthermore, elaborative study of GC-MS analysis was conducted on bioconverted oil extract of DHA to identify the transformation products present in bDHA. The results of this study indicate that the oil extract of bDHA has potential value of industrial significance to control plant pathogenic fungi.

Keywords: Docosahexaenoic acid - Bioconversion - Antifungal activity - Plant pathogens - *Pseudomonas aeruginosa* PR3

AnalÃa Edith PerellÃ^{3, 2}, Maria Virginia Moreno^{1, 2}, Cecilia MÃnaco^{1, 3}, MarÃa Rosa SimÃn⁴ and Cristina Cordo^{1, 3}. (1Centro de Investigaciones de FitopatologÃa (CIDEFI), Facultad de Ciencias Agrarias y Forestales, Universidad Nacional de La Plata, 60 y 119, 1900 La Plata, Provincia de Buenos Aires, Argentina, 2Consejo Nacional de Investigaciones CientÃficas y TÃcnicas (CONICET), Buenos Aires, Argentina, 3ComisiÃn de Investigaciones CientÃficas de la Provincia de Buenos Aires (CIC), La Plata, Argentina, 4Cerealicultura, Universidad Nacional de La Plata, La Plata, Argentina). Biological control of *Septoria tritici* blotch on wheat by *Trichoderma* spp. under field conditions in Argentina. *BioControl*, Volume 54(1) (2009): 113-122

Biological control is an additional tool available for the design of more sustainable control strategies of wheat diseases. *Trichoderma* spp. have previously been used as biocontrol agents to protect wheat plants against leaf spots diseases in Argentina, but the information from field assays is scarce. The effectiveness of four *Trichoderma harzianum* strains and one *T. koningii* strain in reducing the incidence and severity of the leaf blotching of wheat caused by *Septoria tritici* blotch (STB) under two formulation conditions, spore suspension and the coated-seed technique, was studied under field conditions. Significant differences between wheat cultivars, formulation types and growth stages were found. In 2003, at the tillering stage, all of the treatments tested (except SST1 for incidence) effectively reduced the incidence or the severity of the disease compared to the control. Similarly, in 2004, ten of the treatments reduced the severity at tillering. At the heading stage, none of the treatments tested caused a significant decrease of the disease. These results indicated, therefore, that the antagonism was effective at an early stage of the disease only. Comparing both formulations, spraying spore suspension onto leaves and the coated-seed application technique, both were effective in decreasing the disease. Some isolates, such as CST4 and CST2, reduced the incidence value of STB to 40% and the severity value to 70% of the control values applied as coated-seed formulation. On the other hand, isolates T4 and T2 showed the greatest effectiveness for controlling STB, with similar reduction values to that shown by the fungicide (Folicur[®]) application treatment. The results of this study indicated that, although the immediate impact of *Trichoderma* isolates may be seen as reduced incidence and

severity on the first stages of STB, in the long term, the same disease levels as found in untreated sites may be attained. This study also demonstrated that the incorporation of *Trichoderma* as a biocontrol preparation may be a promising step towards reducing STB disease in the field and the levels of fungicide residues in the context of a more integrated approach to the problem.

Keywords: Wheat diseases - Biological control

O. Ajuonu¹, M. Byrne², M. Hill³, P. Neuenschwander¹ and S. Korie¹. (¹Biological Control Centre for Africa, International Institute of Tropical Agriculture (IITA), 08 B.P.09 32 Tri Postal, Cotonou, Benin, ²School of Animal, Plant and Environmental Sciences, University of the Witwatersrand, Private Bag X3, Johannesburg, 2050, South Africa, ³Department of Zoology and Entomology, Rhodes University, P.O. Box 94, Grahamstown, 6140, South Africa). **The effect of two biological control agents, the weevil *Neochetina eichhorniae* and the mirid *Eccritotarsus catarinensis* on water hyacinth, *Eichhornia crassipes*, grown in culture with water lettuce, *Pistia stratiotes*. *BioControl*, Volume 54(1) (2009): 155-162**

We assessed the effect of two biological control agents, the mirid *Eccritotarsus catarinensis* (Carvalho) and the weevil *Neochetina eichhorniae* (Warner), singly or in combination, on the competitive ability of their host plant, water hyacinth, *Eichhornia crassipes* (Mart.) Solms-Laub., grown in a screen house, in competition with another aquatic plant (*Pistia stratiotes* L.). Water hyacinth plant growth characteristics measured included fresh weight, leaf and petiole lengths, number of inflorescences produced, and new shoots. Without herbivory, water hyacinth was 18 times more competitive than water lettuce (across all experimental combinations of initial plant densities), as estimated from fresh weights. Both insect species, singly or in combination, reduced water hyacinth plant growth characteristics. *E. catarinensis* alone was less damaging than the weevil and under normal conditions, i.e., floating water hyacinth, is not expected to increase control of water hyacinth beyond that of the weevil. When combined with the weevil, half the inoculum of weevils and half the inoculum of mirids produced the same growth reduction as the full inoculum of the weevil. Under conditions where the weevils are not effective because water hyacinths are seasonally rooted in mud, the mirid, which lives entirely on leaves, should become a useful additional biological control agent.

Keywords: Competition - *Eccritotarsus catarinensis* - *Neochetina eichhorniae* - Water hyacinth - Water lettuce and weed biological control

Alexander R. Mendoza¹ and Richard A. Sikora¹. (¹Institute for Crop Sciences and Resource Conservation (INRES), Phytopathology in Soil-Ecosystems & Nematology, Bonn, Germany). **Biological control of *Radopholus similis* in banana by combined application of the mutualistic endophyte *Fusarium oxysporum* strain 162, the egg pathogen *Paecilomyces lilacinus* strain 251 and the antagonistic bacteria *Bacillus firmus*. *BioControl*, Volume 54(2) (2009): 263-272**

The biological control efficacy of single or multiple applications of the mutualistic endophyte *Fusarium oxysporum* strain 162, the egg pathogen *Paecilomyces lilacinus* strain 251 and the antagonistic bacteria *Bacillus firmus* toward *Radopholus similis* was investigated in pot trials with banana under glasshouse conditions. *R. similis* was controlled substantially in single and combined applications of *F. oxysporum* with *P. lilacinus* or *B. firmus*. The combination of *F.*

oxysporum and *P. lilacinus* caused a 68.5% reduction in nematode density whereas the individual applications reduced the density by 27.8% and 54.8% over the controls, respectively. Combined application of *F. oxysporum* and *B. firmus* was the most effective treatment in controlling *R. similis* on banana (86.2%), followed by *B. firmus* alone (63.7%). The compatibility of the biocontrol agents, as well the capacity of *F. oxysporum* to colonize banana roots in the absence or presence of *P. lilacinus* was also investigated. *P. lilacinus* did not adversely affect endophytic colonization by *F. oxysporum*. Biological control of *R. similis* in banana can therefore be enhanced via combined applications of antagonists with different modes of action that target different stages in the infection process.

Keywords: Antagonist - Compatibility - Burrowing nematode - *Musa* - Nematode - Root colonization

Jeerapun Worapong^{1, 2} and Gary A. Strobel³. (¹Department of Biotechnology, Faculty of Science, Mahidol University, Rama 6 Rd., Payathai, Bangkok, 10400, Thailand, ²Institute of Science and Technology for Research and Development, Mahidol University, Bhuddhamonthon 4 Rd., Nakornprathom, 73170, Thailand, ³Department of Plant Sciences and Plant Pathology, Montana State University, Bozeman, MT 59717, USA). **Biocontrol of a root rot of kale by *Muscodor albus* strain MFC2. *BioControl*, Volume 54(2) (2009): 301-306**

Pythium ultimum is an oomycetous root rot pathogen that causes significant crop production losses on many crops including kale (*Brassica oleracea*), an economically important vegetable in Thailand. An endophytic fungus from Thailand designated *Muscodor albus* MFC2 controlled *P. ultimum* both in vitro and on kale seedlings grown under outdoor conditions via the production of volatile antibiotics. Ten-day old *M. albus* MFC2 PDA cultures killed *P. ultimum* in vitro. Thoroughly mixing three PDA plates of 10-day old *M. albus* MFC2 into a 500 g mixture of commercial soil and field soil did not adversely affect kale seed germination. The same amount of *M. albus* MFC2 could restore seedling emergence in *P. ultimum* inoculated soil to a level close to that of a non-infested control. In addition, *M. albus* MFC2 did not cause any disease symptoms, but rather seemed to promote the growth of kale in the presence or absence of *P. ultimum* for up to eight weeks after planting.

Keywords: Antagonistic effect - Endophytic biocontrol agent - Fungal fumigant - *Muscodor albus* - *Pythium ultimum* - Root rot of kale

Recep Kotan^{1*}, Neslihan Dikbas² and Hidayet Bostan¹. (¹Atatürk University, Faculty of Agriculture, Department of Plant Protection, Campus, TR-25240-Erzurum, Turkey, ²Ataturk University, Biotechnological Application and Research Center, TR-25240-Erzurum, Turkey. *Corresponding author. E-mail: rkotan@atauni.edu.tr). **Biological control of post harvest disease caused by *Aspergillus flavus* on stored lemon fruits. *African Journal of Biotechnology* Volume 8 (2) (2009): 209–214**

Antagonistic activity of 24 selected bacterial strains detected by previous microbiological studies to *Aspergillus flavus* was tested *in vitro* and *in vivo* conditions. Within 24 strains, only ten strains showed remarkable inhibition zone (6-34 mm) against the pathogen in assays carried out in Petri plates. Both cell suspension and culture filtrates of prominent 10 bacterial strains were also tested in order to control *A. flavus* on lemon fruits cvs Meyer and Interdonato under storage conditions. The cell suspension of nine strains and the culture filtrates of three strains led to

suppression on disease development on lemon fruits. The highest control was obtained by the cell suspension of *Pantoea agglomerans* RK-153. *Erwinia chrysanthemi* RK-67 and *Bacillus subtilis* RK-6 treatments reduced disease severity on both lemon cultivars. Furthermore, both the cell suspension and culture filtrates of *Burkholderia cepacia* strain RK-277 reduced disease severity on only cvs Interdonato, but not on Meyer. There was no significant difference in decay diameters among those treatments, compared to the negative control at 35 days of inoculation. Even other tested strains also slightly reduced disease severity compared to strains determined as efficient ones; disease severity resulted from other strains were statistically significant, compared to negative control. In conclusion, these strains can be used as new biocontrol agents in controlling postharvest decay of citrus fruit.

Keywords: Antibiosis, *Aspergillus flavus* biocontrol, citrus disease, lemon, *Pantoea agglomerans*, postharvest.

Gyoung Hee Kim¹, Myoung Taek Lim¹, Jae-Seoun Hur², Kyu-Jin Yum³ and Young Jin Koh^{1,*}. (¹Department of Plant Medicine, ²Department of Environmental Education, Sunchon National University, Suncheon 540-742, Korea, ³Coenbio Institute of Research and Development, 613 Mega Bldg., SK Technopark, Sangdaewon-Dong, Seongnam 462-120, Korea). **Biological Control of Tea Anthracnose Using an Antagonistic Bacterium of *Bacillus subtilis* Isolated from Tea Leaves. The Plant Pathology Journal, Volume 25(1) (2009): 99-102**

An antagonistic bacterium of *Bacillus subtilis* BD0310 against *Colletotrichum theae-sinensis* was isolated from the phylloplane of tea trees at a tea plantation in Korea. SC (suspension concentrate)-type biofungicide was formulated with use of the antagonist. Cell viability and antifungal activity of *B. subtilis* were maintained in the formulation more than 12 months at room temperature. The antagonist was sensitive only to copper sulfate among the chemical pesticides currently registered for tea trees in Korea. Greenhouse application demonstrated that the biofungicide more effectively controlled the disease in a protective mode than in a curative mode. Field trial showed that alternate applications of the biofungicide and chemical fungicide were more effective in controlling tea anthracnose than single application of the biofungicide or chemical fungicide with less use of chemicals. This study suggests that the biofungicide of *B. subtilis* BD0310 is an effective method for biological control of anthracnose in tea plantations.

Myoung-Hwan Chi, Sook-Young Park and Yong-Hwan Lee* . (Department of Agricultural Biotechnology, Center for Fungal Genetic Resources, and Center for Fungal Pathogenesis, Seoul National University, Seoul 151-921, Korea). **A Quick and Safe Method for Fungal DNA Extraction. The Plant Pathology Journal, Volume 25(1) (2009): 108-111**

DNA-based studies, including cloning and genotyping, have become routine in fungal research laboratories. However, preparation of high-quality DNA from fungal tissue requires much time and labor and is often a limiting step for high-throughput experiments. We have developed a quick and safe (QS) DNA extraction method for fungi. Time efficiency and safety in the QS method were achieved by using plate-grown mycelia as the starting material, by eliminating phenol-chloroform extraction procedures, and by deploying a simple electric grinder. This QS method is applicable not only to a broad range of microbial eukaryotes, including true fungi and oomycetes, but also to lichens and plants.

Fernando Haddad^a, Luiz A. Maffia^a, Eduardo S.G. Mizubuti^a and Hudson Teixeira^a. (^aDepartamento de Fitopatologia, Universidade Federal de Viçosa, Av. P. H. Rolfs s/n, 36570-000 Viçosa, MG, Brazil). **Biological control of coffee rust by antagonistic bacteria under field conditions in Brazil. *Biological Control*, Volume 49(2) (2009): 114-119**

Rust (*Hemileia vastatrix*) is the most important coffee disease in Brazil. Organic coffee production has increased in the country and a research program aimed to develop alternatives to chemicals for disease control was required. Seven bacterial isolates, isolated from organic coffee plantings and selected in greenhouse tests, were evaluated under commercial organic crop conditions in 2005 (Experiment 1) and 2005/2006 (Experiment 2), in Machado, MG, Brazil. Ten treatments consisting of the seven bacterial isolates, copper hydroxide, calcium silicate and water were applied as three or four monthly sprays in Experiment 1 or 2, respectively. Rust severity and incidence were evaluated monthly. In Experiment 1, the sprays started in January when rust incidence was 23.8%, and none of the treatments reduced rust progress significantly. In Experiment 2, the sprays began in November 2005, when rust incidence was approximately 7.5%. There were significant differences ($P < 0.0001$) between treatments regarding maximum incidence and severity (as assessed in June, 2006), the rate of increase of the incidence between November 2005 and June 2006 and for the areas under disease progress curves for both rust incidence and severity. Lower values for these treatments were obtained in the plots treated with copper hydroxide or *Bacillus* sp. isolate B157, and intermediate values with the *Pseudomonas* sp. isolate P286. In a third experiment conducted in 2007 in Ervália, MG, isolates B157 and P286 were also evaluated; isolate B157 reduced rust intensity as effectively as copper hydroxide. Isolate B157 is considered a potential biocontrol agent for coffee rust for organic crop systems in Brazil.

Keywords: *Coffea arabica*; *Hemileia vastatrix*; *Pseudomonas*; *Bacillus*; Biocontrol; Disease management

George A. Bardas^a, Anastasia L. Lagopodi^a, Kalliopi Kadoglidou^b and Katina Tzavella-Klonari^a. (^aLaboratory of Plant Pathology, School of Agriculture, Aristotle University of Thessaloniki, P.O. Box 269, GR541 24 Thessaloniki, Greece, ^bLaboratory of Agricultural Chemistry, School of Agriculture, Aristotle University of Thessaloniki, P.O. Box 251, GR541 24 Thessaloniki, Greece). **Biological control of three *Colletotrichum lindemuthianum* races using *Pseudomonas chlororaphis* PCL1391 and *Pseudomonas fluorescens* WCS365. *Biological Control*, Volume 49(2) (2009): 139-145**

Colletotrichum lindemuthianum is the causal agent of anthracnose, one of the most important diseases of bean worldwide. The rhizobacteria *Pseudomonas chlororaphis* PCL1391 and *Pseudomonas fluorescens* WCS365, known for their biocontrol ability against the tomato pathogen *Fusarium oxysporum* f. sp. *radicis-lycopersici*, were tested *in planta* against three *C. lindemuthianum* races. *Pseudomonas chlororaphis* PCL 1391 in the absence as well as in the presence of the pathogen promoted several plant growth characteristics. The promoting effect was greater regarding certain growth characteristics when this strain was tested in combination with *P. fluorescens* WCS365. On the contrary, treatment with *P. fluorescens* WCS365 resulted in minor differentiations in the growth characteristics. Treatment with *P. chlororaphis* PCL1391 resulted in best biocontrol of anthracnose, while *P. fluorescens* WCS365 showed no significant difference compared to the positive control. The combined bacterial treatment did not differ from the treatment with *P. chlororaphis* PCL1391 alone. Colonization experiments under gnotobiotic conditions showed that *P. chlororaphis* PCL1391 and *P. fluorescens* WCS365 are

both excellent colonizers of bean roots. Their combined treatment resulted in increased total bacterial populations on the root tips and reduction of the *P. fluorescens* WCS365 population. When tested against the three races of the pathogen *in vitro*, *P. chlororaphis* PCL1391 reduced pathogen growth, sporulation, and conidial germinability. Similar results were obtained when both bacteria were used in combination. In contrast, *P. fluorescens* WCS365 applied alone did not affect any of these characteristics. It was assumed that phenazine-1-carboxamide produced by *P. chlororaphis* PCL1391 was the crucial factor for the *in vitro* activity of this strain. This hypothesis was supported by the absence of fungal growth over phenazine-1-carboxamide on a TLC plate seeded with *C. lindemuthianum* spores. In conclusion, *P. chlororaphis* PCL1391 alone or combined with *P. fluorescens* WCS365 can be a potential factor in integrated control systems against bean anthracnose in Greece.

Keywords: Bean anthracnose; Colonization; Integrated control; Phenazine-1-carboxamide; PGPR

Joeke Postma^a, Luc H. Stevens^a, Gerrie L. Wieggers^a, Evert Davelaar^a and Els H. Nijhuis^a. (^aPlant Research International, 6700 AA Wageningen, P.O. Box 16, The Netherlands). **Biological control of *Pythium aphanidermatum* in cucumber with a combined application of *Lysobacter enzymogenes* strain 3.1T8 and chitosan. *Biological Control*, Volume 48(3) (2009): 301-309**

Pythium aphanidermatum (Edson) Fitzp., causing root and crown rot in cucumber, was successfully managed by *Lysobacter enzymogenes* strain 3.1T8. Greenhouse experiments were performed with cucumber plants grown in rockwool blocks up to 5 weeks with a recirculated nutrient solution. Application of *L. enzymogenes* 3.1T8 in combination with chitosan (the deacetylated derivative of chitin) reduced the number of diseased plants by 50–100% in four independent experiments relative to the *Pythium* control. Application of chitosan or the bacterial inoculant alone was not effective. Washed bacterial cells plus chitosan inhibited *Pythium*-induced disease, but the supernatant without bacterial cells combined with chitosan was not effective. The most effective and convenient type of commercially available chitosan was selected. Chitosan disappeared from the hydroponic system within 24 h after application, which we attribute to enzyme expression of *L. enzymogenes* 3.1T8 induced by the exposure to chitosan. Plate counts of the nutrient solution on a general bacterial medium showed the dominance of the inoculated strain, and an increased bacterial population growing on chitin and chitosan as single carbon source. The population density of *L. enzymogenes* 3.1T8 on the cucumber roots was investigated with a strain specific real-time TaqMan PCR. Highest chitosan concentrations applied (0.1 and 0.03 g/plant) resulted in the highest numbers of *L. enzymogenes* 3.1T8 present on roots; i.e. 10^8 – 10^9 cells/g root. Substantially higher numbers of bacterial cells were observed by scanning electron microscopy after application of chitosan; no morphological or other qualitative differences were found. The results indicate that addition of chitosan enhanced the biocontrol efficacy of *L. enzymogenes* 3.1T8; either chitosan serves as C- and N-source for the antagonist, induces antagonistic gene expression, or both.

Keywords: Biological control; *Lysobacter enzymogenes*; Chitosan; Synergistic effect; Quantitative PCR; Root colonization

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Bacillus mycoides isolate BmJ (BmJ) and *Bacillus mojavensis* isolate 203-7 (203-7) were tested in the greenhouse for their ability to control *Glomerella cingulata* var. *orbiculare* the causal agent of anthracnose of cucumber by induced systemic acquired resistance (SAR). BmJ and 203-7 delayed disease onset and reduced total (43% and 56%) and live spore production (38% and 49%) per mm² of lesion area when used to induce SAR in cucumber. 203-7 also reduced lesion diameter. Induction by *G. cingulata* conidia resulted in delayed disease onset, reduction of number of lesions per leaf and lesion diameter. Assays of cucumber apoplastic proteins extracted 6 days after induction showed that BmJ increased β -glucanase activity by 135%, and 203-7 increased β -glucanase activity by 72% and peroxidase activity by 79% when compared to the water control. Acibenzolar-*S*-methyl induced the highest ($P = 0.05$) levels of chitinase (950%) and peroxidase (420%) activity compared to water controls. Field experiments (2004 and 2005) evaluated applications of BmJ and fungicides for the control of anthracnose in cucumber (var. 'General Lee') and cantaloupe (var. 'Athena'). BmJ was compared to full and half labeled rate alternate applications of azoxystrobin and chlorothalonil, and BmJ with half rate of azoxystrobin and chlorothalonil. BmJ applied seven days before inoculation reduced disease severity by 41% in cucumber in 2004 and by 24–21% in cantaloupe for both years compared to water controls which was statistically equal to the fungicide treatments. The full and half rate fungicide program provided 97–37% disease reduction compared to water controls. BmJ applied one week before inoculation significantly reduced AUDPC ($P = 0.05$) in cucumber compared to the water control in 2004 on cantaloupe for both years while the full and half rate fungicide program were equivalent and provided the lowest AUDPC. No yield reduction was noted as a result of the disease or treatment for either cantaloupe or cucumber.

Keywords: *Bacillus mycoides*; *Bacillus mojavensis*; *Glomerella cingulata* var. *orbiculare*; Anthracnose of cucumber; Anthracnose of melon; Biological control; Systemic acquired resistance; *Cucumis sativus*; *Cucumis melo*

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Bellyache bush, *Jatropha gossypifolia* L., is a serious weed of northern Australia. *Agonosoma trilineatum* (F.) is an insect from tropical America released in Australia in 2003 as a biological control agent against bellyache bush. It feeds on seeds and has the potential to reduce seed

production, thereby potentially reducing the rate of spread and recruitment. To test the host specificity of *A. trilineatum*, four biological responses to host plant species were determined: development of nymphs, oviposition preferences, adult feeding and frequency of mating. Development of nymphs to adults and adult feeding only occurred on three *Jatropha* spp. These species also supported mating and oogenesis but only *J. gossypifolia* was accepted for oviposition. Mating did not occur in the presence of other plant species. The evidence indicates that there is little risk associated with the release of this insect species in Australia and probably other countries where this weed is a problem. The probability of this insect expanding its host range is low because multiple aspects of the biology would need to change simultaneously. *A. trilineatum* was released in Australia between 2003 and 2007. A Climex model indicated that coastal areas of Queensland and the Northern Territory would be climatically most suitable for this insect.

Keywords: Host specificity; Host range; Biological control of weeds; Climate matching

Hongyin Zhang^a, Lei Wang^{a, b}, Longchuan Ma^a, Ying Dong^a, Song Jiang^a, Bin Xu^a and Xiaodong Zheng^c. (^aCollege of Food and Biological Engineering, Jiangsu University, Xuefu Road 301, Jiangsu 212013, Zhenjiang, P.R. China, ^bCollege of Applied Technology, Ludong University, Yantai 264025, Shandong, P.R. China, ^cDepartment of Food Science and Nutrition, Zhejiang University, Hangzhou 310029, Zhejiang, P.R. China). **Biocontrol of major postharvest pathogens on apple using *Rhodotorula glutinis* and its effects on postharvest quality parameters. Biological Control, Volume 48(1) (2009): 79-83**

The biocontrol activity of *Rhodotorula glutinis* on gray mold decay and blue mold decay of apple caused by *Botrytis cinerea* and *Penicillium expansum*, respectively, was investigated, as well as its effects on postharvest quality of apple fruits. The results show there was a significant negative correlation between concentrations of the yeast cells and the disease incidence of the pathogens. The higher concentration of the *R. glutinis*, the better effect of the biocontrol capacity. At concentrations of *R. glutinis* 1×10^8 CFU ml⁻¹, the amount of gray mold decay was completely inhibited after 5 days incubation at 20 °C, after challenge with *B. cinerea* spores suspension of 1×10^5 spores ml⁻¹; While the blue mold decay was completely inhibited at concentrations of 5×10^8 CFU ml⁻¹, at challenged with *P. expansum* spores suspension of 5×10^4 spores ml⁻¹. These results demonstrated that the efficacy of *R. glutinis* in controlling of gray mold decay of apples was better than the efficacy of controlling blue mold. *R. glutinis* within inoculated wounds on apples increased in numbers at 20 °C from an initial level of 9.5×10^5 CFU per wound to 2.24×10^7 CFU at 20 °C after 1 day. The highest population of the yeast was recovered 4 days after inoculation, the yeast population in wounds increased by 56.9 times. After that, the population of the yeast began to decline very slowly. *R. glutinis* significantly reduced the incidence of natural infections on intact fruit from 75% in the control fruit to 28.3% after 5 days at 20 °C, and from 58.3 to 6.7% after 30 days at 4 °C followed by 4 days at 20 °C. *R. glutinis* treatment had no deleterious effect on quality parameters after 5 days at 20 °C or after 30 days at 4 °C followed by 4 days at 20 °C.

Keywords: Apple; Gray mold; Blue mold; Postharvest decay; Biocontrol; *Rhodotorula glutinis*; Quality parameters

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Eucharitids are specialized parasitoids of ants. The biology, life cycle and chemical ecology are known for a number of species, but the study of the impact of eucharitid wasps upon their ant hosts has been seldom addressed. Here, we determine the prevalence of the parasitism of two sympatric *Kapala* species upon a population of the neotropical ant *Ectatomma ruidum*, along a 12-month sampling period. Adult and immature parasitoids were present in the nests all year round, and several cases of superparasitism were observed. Parasitism varied strongly among the nests for any collecting date and among collecting dates, but the prevalence of *Kapala* parasitoids increased significantly during the rainy season, and the probability for a nest of being parasitized was positively correlated with colony size, particularly with cocoon number. At the population scale, more than 28% of all *E. ruidum* pupae produced during the ant reproductive and dispersal period (June) were infested. Our results are discussed from the point of view of the impact of these parasitoids on the colonies of *E. ruidum*, a potential biocontrol agent in coffee and cocoa plantations in southeastern Mexico.

Keywords: Prevalence of parasitism; Host–parasitoid interaction; Negative impact; *Kapala izapa*; *Kapala iridicolor*; *Ectatomma ruidum*

P.C. Abhilash^a and Nandita Singh^a. (^aEco-Auditing Group, National Botanical Research Institute, Council of Scientific and Industrial Research, Rana Pratap Marg, Lucknow 226001, Uttar Pradesh, India). **Pesticide use and application: An Indian scenario. Journal of Hazardous Materials, Volume 165(1-3) (2009): 1-12**

Agricultural development continues to remain the most important objective of Indian planning and policy. In the process of development of agriculture, pesticides have become an important tool as a plant protection agent for boosting food production. Further, pesticides play a significant role by keeping many dreadful diseases. However, exposure to pesticides both occupationally and environmentally causes a range of human health problems. It has been observed that the pesticides exposures are increasingly linked to immune suppression, hormone disruption, diminished intelligence, reproductive abnormalities and cancer. Currently, India is the largest producer of pesticides in Asia and ranks twelfth in the world for the use of pesticides. A vast majority of the population in India is engaged in agriculture and is therefore exposed to the pesticides used in agriculture. Although Indian average consumption of pesticide is far lower than many other developed economies, the problem of pesticide residue is very high in India. Pesticide residue in several crops has also affected the export of agricultural commodities in the last few years. In this context, pesticide safety, regulation of pesticide use, proper application technologies, and integrated pest management are some of the key strategies for minimizing human exposure to pesticides. There is a dearth of studies related to these issues in India. Therefore, the thrust of this paper was to review the technology of application of pesticides in India and recommend future strategies for the rational use of pesticides and minimizing the problems related to health and environment.

Keywords: Pesticide; Agriculture; Application technology; Residue

Biodegradation

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Laboratory incubation experiments were carried out to assess the potential of methanotrophic culture for degrading TCE. Measurements of the growth rate and TCE degradation showed that the methanotrophs not only grew in presence of TCE but also degraded TCE. The rate of TCE degradation was found to be 0.19 ppm h⁻¹. The reverse transcriptase-PCR test was conducted to quantify expression of *pmoA* and *mmoX* genes. RT-PCR revealed expression of *pmoA* gene only. This observation provides evidence that the *pmoA* gene was functionally active for pMMO enzyme during the study. The diversity of the methanotrophs involved in TCE degradation was assessed by PCR amplification, cloning, restriction fragment length polymorphism and phylogenetic analysis of *pmoA* genes. Results suggested the occurrence of nine different phylotypes belonging to Type II methanotrophs in the enriched cultures. Out of the nine, five clustered with, genera *Methylocystis* and rest got clustered in to a separate group.

Keywords: Methanotrophs; TCE; RT-PCR; pMMO; Bioremediation

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In this study, the evolution of the most important parameters (temperature, pH, electrical conductivity, total organic carbon, total nitrogen, and C/N ratio) describing the composting process of olive oil husk with other organic wastes was investigated. Four windrows for obtaining two mixed wastes composts (MWCs) and two green wastes composts (GWCs) were prepared.

All the raw materials used showed appropriate physical and chemical properties for composting process. The total organic carbon values of the final composts were suitable for agricultural purpose and in particular two of them (one MWC and one GWC) showed an increase of 47.6% and 40.3% in respect to the minimum levels established by the Italian legislation. After the biodegradation the C/N ratio could be considered satisfying for ready-to-use compost in three of the four windrows. The Ni and Pb concentrations did not overcome the Italian law limits in all windrows, while the Zn content was higher than the limit value only in two windrows (one of both MWC and GWC composts).

The findings highlighted that among the four composting processes, the best general results were found for one of the two GWCs produced.

Keywords: Olive oil husk; Composting; Mixed and green wastes composts; Heavy metals content

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Lab-scale batch studies were conducted to determine the biodegradability of oil associated with oily sludge from a steel mill using two microbial cultures enriched in the laboratory. After 60 days of biodegradation the residual oil content in mill sludge was reduced from 4.5–5% to 2.7–3.0%, corresponding to 40–45% loss with respect to initial. The rate of degradation was different for the two enrichment cultures studied. Significant loss of oil was observed in the un-inoculated controls while loss in the azide killed controls was negligible. Bioavailability limitations and the presence of structurally complex high molecular weight hydrocarbons in lubricating oil are responsible for the slow rate of degradation. Significant loss of oil in un-inoculated controls indicated the presence of indigenous microorganisms in oily mill sludge. The association of biomass with sludge solids and presence of a high level of residual oil may adversely affect the recyclability of iron-fines associated with the sludge.

Keywords: Lubricating oil; Oily sludge; Bioavailability; Iron-fines; Biological treatment

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A systematic study on the anaerobic degradability of a series of starch:polyvinyl alcohol (TPS:PVOH) blends was performed to determine their fate upon disposal in either anaerobic digesters or bioreactor landfills. The aims of the study were to measure the rate and extent of solubilisation of the plastics. The extent of substrate solubilisation on a COD basis reached 60% for a 90:10 (w/w) blend of TPS:PVOH, 40% for 75:25, 30% for 50:50 and 15% for PVOH only. The rate of substrate solubilisation was most rapid for the 90:10 blend (0.041 h^{-1}) and decreased with the amount of starch in the blend in the following order 0.034 h^{-1} (75:25); 0.023 h^{-1} (50:50). The total solids that remained after 900 h were 10 wt.% (90:10); 23 wt.% (75:25); 55 wt.% (50:50); 90 wt.% (0:100). Starch containing substrates produced a higher concentration of volatile fatty acids (VFAs) and biogas, compared to the 0:100 substrate. The major outcome was that PVOH inhibited the degradation of the starch from the blend.

Keywords: Anaerobic digestion; Degradation; Carbohydrates; Starch; Polyvinyl alcohol

Jinyou Shen^a, Rui He^a, Hongxia Yu^a, Lianjun Wang^a, Jianfa Zhang^a, Xiuyun Sun^a, Jiansheng Li^a, Weiqing Han^a and Lu Xu^a. (^aSchool of Chemical Engineering, Nanjing University of Science and Technology, 200 Xiaolingwei Street, Nanjing 210094, Jiangsu Province, China). **Biodegradation of 2,4,6-trinitrophenol (picric acid) in a biological aerated filter (BAF). Bioresource Technology, Volume 100(6) (2009): 1922-1930**

This study demonstrated the microbial purification of a model wastewater containing 2,4,6-trinitrophenol (TNP), which was carried out in a continuously working biological aerated filter (BAF). The main emphasis was on the operating performance of the reactor as a function of the pollution load. TNP was degraded at a maximum volumetric removal rate of 2.53 g TNP/L d, with low residual COD and TNP concentration. Overloading of TNP inhibited the nitrite-oxidizing activity, resulting in poor TNP degradation performance in the BAF system. The inhibition depended on some factors, such as influent concentrations and flow rates of the influent. It is assumed that nitrite-oxidizing occurred spontaneously during TNP degradation in the BAF system, could have significant influence on TNP wastewater treatment. One year after the reactor start-up, the dominance of *Rhodococcus*, which was initially inoculated in the reactor, was confirmed by analysis of 16S rDNA sequence of the PCR products separated by DGGE.

Keywords: Biodegradation; 2,4,6-Trinitrophenol (TNP); Biological aerated filter (BAF); PCR-DGGE; Nitrite-oxidizing

Chunli Zheng^a, Jiti Zhou^a, Jing Wang^a, Baocheng Qu^a, Jing Wang^a, Hong Lu^a and Hongxia Zhao^a. (^aSchool of Environmental and Biological Science and Technology, Dalian University of Technology, Dalian 116024, Liaoning Province, China). **Aerobic degradation of 2-picolinic acid by a nitrobenzene-assimilating strain: *Streptomyces* sp. Z2. Bioresource Technology, Volume 100(6) (2009): 2082-2084**

Streptomyces sp. Z2 was isolated from nitrobenzene contaminated activated sludge, which utilized nitrobenzene as a sole source of carbon, nitrogen, and energy under aerobic condition. It was found that besides nitrobenzene strain Z2 can degrade 2-picolinic acid. Strain Z2 completely degraded 2-picolinic acid with initial concentration of 500 mg/L, 1000 mg/L, 1500 mg/L, 2000 mg/L, 2500 mg/L, and 3000 mg/L within 36 h, 50 h, 72 h, 100 h, 136 h, and 180 h, respectively. Kinetics of 2-picolinic acid degradation was described using the Andrews equation. The kinetic parameters were as follows: $q_{\max} = 3.81 \text{ h}^{-1}$, $K_s = 83.10 \text{ mg/L}$, and $K_i = 252.11 \text{ mg/L}$. During the biodegradation process, Z2 transformed 2-picolinic acid into a product which was identified as 6-hydroxy picolinic acid by UV-vis spectrometry, ¹H nuclear magnetic resonance spectroscopy, and mass spectrometry. 6-Hydroxy picolinic acid was then cleaved and mineralized with release of ammonia.

Keywords: 2-Picolinic acid; Nitrobenzene; Degradation; 6-Hydroxy picolinic acid; *Streptomyces*

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The aim of this work was the study of poly- β -hydroxybutyrate (PHB) formation and degradation in a sequencing batch biofilm reactor (SBBR). The SBBR was operated in cycles comprising three individual phases: mixed fill, aeration and draw. A synthetic substrate solution with acetate and ammonium was used.

PHB was formed during the aeration phase immediately after acetate depletion, and was subsequently consumed for biomass growth, owing to the high oxygen concentration in the reactor. It was observed a combination of suspended and biofilm growth in the SBBR with predominance of the fixed form of biomass (506 Cmmol and 2102 Cmmol, respectively). Maximum PHB fraction of suspended biomass (0.13 Cmol/Cmol) was considerably higher than that of biofilm (0.01 Cmol/Cmol). This may possibly be explained by a combination of two factors: lower mass transfer limitation of acetate and higher fraction of heterotrophs in suspended biomass compared to the ones of biofilm.

Keywords: Poly- β -hydroxybutyrate (PHB); Sequencing batch biofilm reactor (SBBR); Nitrogen removal

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The biodegradation of Congored, a toxic azo dye, was studied by using a hybrid technique involving sonolysis as pretreatment followed by biological treatment. The experiments were carried out with and without pretreatment using dye solution as a sole source of nutrition with an isolated and acclimatized strain of *Bacillus* sp. obtained from tannery industry effluent. The pretreatment time was varied as 30, 60, 90, 120, 150 and 180 min and then the pretreated dye solution was subjected to biological treatment. The effectiveness of pretreatment was compared with the results of biological degradation of non pretreated Congored and the results showed that the pretreatment improved the efficiency of the biodegradation of Congored. During the biological degradation, the increase in initial dye concentration decreased the decolorization rate and at high concentrations (1500 and 2000 mg/l), the inhibition was observed. The optimum pH and temperature were determined to be 7.0 and 37 °C, respectively. The data obtained through biodegradation experiments were fitted with five different kinetic models and the results were analyzed.

Keywords: *Bacillus* sp.; Congored; Biodegradation; Sonolysis; Kinetics

Kiyohiko Nakasaki^a, Le Thi Hong Tran^a, Yoshito Idemoto^a, Michiharu Abe^a and Analiza Palenzuela Rollon^b. (^aDepartment of Materials Science and Chemical Engineering, Shizuoka University, 3-5-1, Johoku, Naka-ku, Hamamatsu 432-8561, Japan, ^bDepartment of Chemical Engineering, University of the Philippines, Diliman, 1101 Quezon City, Philippines). **Comparison of organic matter degradation and microbial community during thermophilic composting of two different types of anaerobic sludge. Bioresource Technology, Volume 100(2) (2009): 676-682**

Changes in organic matter degradation and microbial communities during thermophilic composting were compared using two different types of anaerobic sludge, one from mesophilic methane fermentation, containing a high concentration of proteins (S-sludge), and the other from thermophilic methane fermentation, containing high concentrations of lipids and fibers (K-sludge). The difference in the organic matter degradation rate corresponded to the difference in the organic matter constituents; the CO₂ evolution rate was greater in the composting of S-sludge than of K-sludge; moreover, the NH₃ evolution resulting from the protein degradation was especially higher in the composting of S-sludge. Then the differences in the microbial communities that contributed to each composting were determined by the PCR-DGGE method. *Ureibacillus* sp., which is known as a degrader with high organic matter degradation activity, was observed during the composting of S-sludge, whereas *Thermobifida fusca*, which is a well known thermophilic actinomycete that produces enzymes for lignocellulose degradation, were observed during the composting of K-sludge.

Keywords: Anaerobic sludge; Composting; Microbial community structure; *Ureibacillus* sp.; *Thermobifida fusca*

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A simple technique, REMI (restriction enzyme-mediated integration), was used to construct transformants of *Trichoderma atroviride* with improved capability of degrading organophosphate pesticide dichlorvos. Linearized DNA of plasmid pV2 bearing the hygromycin B phosphotransferase (*hph*) gene was inserted into chromosomes of wild strain T23 and transformation was confirmed by PCR and Southern blot analysis, respectively. Of 247 transformants, 76% showed improved dichlorvos degradation ability as compared to the parent strain T23 based on the least significant difference (LSD) test at $p = 0.01$. Among them, 8 transformants exhibited 30% higher in degradation rate than the parent isolate. The highest dichlorvos degradation rate of the transformants was up to 96%. This study provided an effective approach for improving organophosphate pesticide-degrading capability of *T. atroviride*.

Keywords: REMI; *Trichoderma*; Organophosphate pesticide; Dichlorvos; Biodegradation

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Interdisciplinary Science & Technology (NIST) (Formerly Regional Research Laboratory), CSIR, Trivandrum, 695019, India, ⁵Biotechnology Process, Department of Chemical Engineering, Universidade Federal do Paraná, Av. Cel. Francisco H. dos Santos, 210, Jardim das Américas, Curitiba, PR, Brazil). Phytodegradation Potential of *Erythrina crista-galli* L., Fabaceae, in Petroleum-Contaminated Soil. *Applied Biochemistry and Biotechnology*, Volume 157(1) (2009): 10-22

This work aimed at investigating both the tolerance and the phytodegradation potential of *Erythrina crista-galli* L. in petroleum-contaminated soil. It consisted in analyzing *E. crista-galli* germination, surviving, growth, and development when cultivated at different contaminant concentrations and pollutant degradation rates. This specimen was selected because it presented a special behavior among others also exposed to petroleum in an accident that occurred in the Araucaria region (south of Brazil), resulting in a four-million-liter oil spill. The experiment was carried out in a greenhouse containing non-contaminated soil (NCS), vegetated contaminated soil (VCS), and non-vegetated contaminated soil (NVCS) at the following petroleum concentrations: 25 g kg⁻¹ (VCS-25), 50 g kg⁻¹ (VCS-50), and 75 g kg⁻¹ (VCS-75). After 60 days, the soil samples were analyzed by gas chromatography. Germination was more and more evident as higher petroleum concentrations were observed. The surviving rates of groups NCS, VCS-25, VCS-50, and VCS-75 were 64%, 70%, 61%, and 96%, respectively. The VCS group growth was reduced when compared to the control group (NCS). The individuals exposed to petroleum pollution presented differences in the anatomic structure of their roots when compared to the NCS group. It was observed that the petroleum degradation rate was higher for VCS group than for NVCS. *E. crista-galli* is potentially recommended for petroleum-contaminated soils because of its positive association in the presence of contamination.

Keywords Phytodegradation - Petroleum - *Erythrina* - Root - Fabaceae

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Forty six bacterial isolates able to grow on crude oil were isolated from various hydrocarbon-contaminated sites in Kuwait. The extent of crude oil degradation varied over a wide range (1–87%) among the isolates. Isolates were predominantly Gram-positive bacteria (79% of total isolates) belonging to the genera *Bacillus* (93%) and *Paenibacillus* (7%). Among the few Gram-negative isolates were from the genera *Acinetobacter*, *Alcaligenes*, *Klebsiella*, *Burkholderia*, *Pseudomonas*, and *Williamsia*. Analyses of their cell-surface hydrophobicity (CSH) by various methods equally showed a wide variation among the isolates. About 74% of isolates that degraded significant amounts of crude oil (>40% degradation) possessed high level of CSH, while 58% of all the isolates exhibited high levels of CSH. Statistical analyses showed significantly high correlation between the ability to degrade crude oil and CSH. The ability of the isolates to bind to polystyrene and salt-aggregation test as measures of CSH were more strongly correlated with hydrocarbon-degrading ability than adherence to hydrocarbons.

Keywords: Bacterial cell-surface hydrophobicity; Hydrocarbon-degradation

Franciscon Elisangela^a, Zille Andrea^b, Dias Guimaro Fabio^a, Ragagnin de Menezes Cristiano^a, Durrant Lucia Regina^a and Cavaco-Paulo Artur^c. (^aCampinas State University, Department of Food Science, 13083-970 Campinas, São Paulo, Brazil, ^bIBMC – Instituto de Biologia Molecular e Celular, Universidade do Porto, Portugal, ^cUniversity of Minho, Department of Textile Engineering, 4800-058 Guimarães, Portugal). **Biodegradation of textile azo dyes by a facultative *Staphylococcus arlettae* strain VN-11 using a sequential microaerophilic/aerobic process. International Biodeterioration & Biodegradation, Volume 63(3) (2009): 280-288**

A facultative *Staphylococcus arlettae* bacterium, isolated from an activated sludge process in a textile industry, was able to successfully decolourize four different azo dyes under microaerophilic conditions (decolourization percentage >97%). Further aeration of the decolourized effluent was performed to promote oxidation of the degradation products. The degradation products were characterized by FT-IR and UV-vis techniques and their toxicity with respect to *Daphnia magna* was measured. The amine concentrations as well as the total organic carbon (TOC) levels were monitored during the biodegradation process. The presence of aromatic amine in the microaerophilic stage and its absence in the aerobic stage indicated the presence of azoreductase activity and an oxidative biodegradation process, respectively. TOC reduction was ~15% in the microaerophilic stage and ~70% in the aerobic stage. The results provided evidence that, using a single *Staphylococcus arlettae* strain in the same bioreactor, the sequential microaerophilic/aerobic stages were able to form aromatic amines by reductive breakdown of the azo bond and to oxidize them into non-toxic metabolites.

Keywords: Azo dyes; *Staphylococcus arlettae*; Biodegradation; Textile effluents; Aromatic amines

Renato N. Montagnolli^a, Paulo R.M. Lopes^a and Ederio D. Bidoia^a. (^aDepartment of Biochemistry and Microbiology, IB, State University of São Paulo (UNESP), Av. 24 A, no. 1515, 13506-900 Rio Claro, São Paulo, Brazil). **Applied models to biodegradation kinetics of lubricant and vegetable oils in wastewater. International Biodeterioration & Biodegradation, Volume 63(3) (2009): 297-305**

Bioremediation technologies are used in order to remove pollutants from the environment in a safe, economical and harmless way during the treatment of waste, especially with the use of techniques such as biodegradation. A lubricant and vegetable oil contaminated water sample was studied in order to evaluate the biodegradability of different types of oils, considering the relevance of the obtained data in the bioremediation procedures. The objective of this paper is to use respirometry technique as a biodegradation process data source, and then apply to the obtained data the experimental design of mathematical models to characterize and determinate how the different types of oils are capable of affecting the parameters in biodegradation kinetics. The kinetics was then evaluated through selected models with a reasonable fit to experimental data. The Bartha and Pramer respirometer is used as a method to accurately measure the CO₂ formation in the organic compounds degradation by microorganisms. Therefore, the difference in biodegradation efficiency process is compared in the different groups of oils using mathematical models fitting the obtained data for the kinetics of biodegradation. The results demonstrated that used lubricant automotive oils are more susceptible to the biodegradation process, since their molecular structures had already been altered after use. In general,

automotive lubricant oils shown better performance in biodegradation than vegetable oils. The models proposed for the obtained data in each of these assays demonstrated that vegetable oils biodegradation rate tends to decrease faster and end sooner than the automotive oils. Also, the modeling predicted that higher rates of biodegradation and total CO₂ production are to be expected in automotive lubricant oils rather than vegetable oils.

Keywords: Biodegradation; Kinetic model; Lubricant oil; Vegetable oil; Respirometry

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Hydrocarbon biodegradation in clayed and weathered polluted soils is a challenge; thus the aim of the present study was to determine the best experimental conditions that improve the hydrocarbon biodegradability in clayed and weathered polluted soils, modifying the nitrogen and phosphorous content considering the C:N:P ratio and the water content as a percentage of the water-holding capacity of the soil. Biodegradation tests were performed in microcosms containing 20 g of dry soil at 30 °C. A uniform-precision central composite design of second order with three levels was used to assess the effect of nutrient and water content adjustment on the hydrocarbon degradation rate, total carbon consumption, and microbial activity. The results showed that the water-holding capacity corresponding to 350% and a C:N:P ratio of 100:7.5:0.66 were the best experimental conditions for obtaining the highest hydrocarbon degradation rate (1145 mg TPH kg⁻¹ dry soil day⁻¹), whereas the hydrocarbon degradation rate in a non-stimulated control was only 129 mg TPH kg⁻¹ dry soil day⁻¹. Water content was the factor that showed the highest significant effect ($p \leq 0.05$) on the hydrocarbon degradation rate. The results of the present study allowed the achievement of the best experimental conditions that enhance hydrocarbon biodegradability in clayed and weathered polluted soils. Also, these conditions are proposed for use as a biodegradability assay.

Keywords: Drilling muds; Native microorganisms; Nutrient stimulation; Water content; Assay

Astrid Michaelsen^{a, 1}, Guadalupe Piñar^b, Mariasanta Montanari^c and Flavia Pinzari^c. (^aDepartment of Microbial Ecology, University of Vienna, Althanstrasse 14, 1090 Vienna, Austria, ^bInstitute of Applied Microbiology, Department of Biotechnology, Universität für Bodenkultur, Muthgasse 18, 1190 Vienna, Austria, ^cICRCPAL – Istituto Centrale per il Restauro e la Conservazione del Patrimonio Archivistico e Librario, Laboratorio di Biologia, Ministero per i Beni e le Attività Culturali, Via Milano, 76, 00184 Rome, Italy). **Biodeterioration and restoration of a 16th-century book using a combination of conventional and molecular techniques: A case study. International Biodeterioration & Biodegradation, Volume 63(2) (2009): 161-168**

In this paper we deliver a report on the study of microbiological damage found on the pages of a 16th-century book. Our aim is to describe the procedures needed to ensure a conservative approach to the restoration of valuable books and objects of art made from, or supported on, paper. The techniques employed to evaluate and describe the damage observed, as well as the organisms responsible for biodeterioration, are discussed. A range of sampling techniques and instruments were utilised, including swabs and adhesive tape. Conventional methods, such as classic culturing and the direct microscopic observation of sampled material, were coupled with DNA-fingerprinting and phylogenetic analysis. We postulated that the purple stains which migrate through the pages with a felted consistency (Fig. 2), based on all the information obtained using traditional and molecular means, were caused by a cellulolytic fungus producing purple essudates, characterised by echinated conidia and Hülle cells. These elements were consistent with the discovery of both *A. versicolor* and *A. nidulans* using molecular techniques.

Keywords: Paper; Fungi; Biodeterioration; DNA-fingerprints; Phylogenetic identification; Cultural heritage; Restoration

Le Thi Nhi-Cong^a, Annett Mikolasch^a, Hans-Peter Klenk^b and Frieder Schauer^a. (^aErnst-Moritz-Arndt-University Greifswald, Institute for Microbiology, F.L. Jahnstr. 15A, 17487 Greifswald, Germany, ^bDepartment of Microbiology, DSMZ – German Collection of Microorganisms and Cell Cultures, Inhoffenstraße 7B, 38124 Braunschweig, Germany). **Degradation of the multiple branched alkane 2,6,10,14-tetramethyl-pentadecane (pristane) in *Rhodococcus ruber* and *Mycobacterium neoaurum*. International Biodeterioration & Biodegradation, Volume 63(2) (2009): 201-207**

Pristane, a highly branched hydrocarbon that also contains *iso*-branched termini, was used as a substrate for several alkane-metabolizing bacteria. *Rhodococcus ruber* and *Mycobacterium neoaurum* were able to utilize pristane for growth effectively. The intermediates produced by these bacteria during incubation with pristane were analyzed by gas chromatography (GC) and gas chromatography/mass spectra (GC/MS). The products revealed as products of 4-methyl pentanoic acid; methyl butanedioic acid; 2-methyl pentadioic acid; methyl propanedioic acid; 4-methyl heptanedioic acid; and 2,6,10,14-tetramethyl-pentadecan-3-one were detected in *M. neoaurum* cultures. In *R. ruber*, methyl butanedioic acid; 2-methyl pentadioic acid; 4,8-dimethylnonanoic acid, 4-methyl heptanedioic acid; 2,6,10-trimethylundecanoic acid; 3,7-dimethyl decanedioic acid; and 2,6,10,14-tetramethyl-pentadecan-3-one were detected. The occurrence of these intermediates showed that pristane could be catabolized not only via mono- but also by a di-terminal oxidation pathway. Furthermore, the presence of 2,6,10,14-tetramethyl-pentadecan-3-one; 3,7-dimethyldecandioate; and 2-methylbutandioate established a third pathway initiated by sub-terminal oxidation at the third carbon atom of pristane. Novel intermediates detected suggest simultaneous sub-terminal and di-terminal oxidation pathways.

Keywords: Hydrocarbons; Pristane; *Rhodococcus ruber*; *Mycobacterium neoaurum*; Sub-terminal oxidation

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conditions by soil fungi. *International Biodeterioration & Biodegradation*, Volume 63(2) (2009): 224-229

Soil fungi were studied regarding their ability to degrade polycyclic aromatic hydrocarbons (PAHs) and produce ligninolytic enzymes under microaerobic and very-low-oxygen conditions. Several PAHs were used as substrates for soil fungi under microaerobic and very-low-oxygen conditions. Activities of lignin-peroxidase, manganese-peroxidase, and laccase were monitored over a 30-day period. PAH degradations were analyzed using C₁₈ reversed-phase HPLC. Low-molecular-weight PAHs (LMW-PAHs, 2–3 rings) were degraded most extensively by *Aspergillus* sp., *Trichocladium canadense*, and *Fusarium oxysporum*. When growing on high-molecular-weight PAHs (HMW-PAHs, 4–7 rings), the highest degradations were reached by *T. canadense*, *Aspergillus* sp., *Verticillium* sp., and *Achremonium* sp. In this study, these fungi revealed a great capability to degrade a broad range of PAHs under low-oxygen conditions. In addition, lignolytic enzyme activities were observed during fungal growth on these compounds. These results suggest fungi have the ability to bioremediate PAH-contaminated soils and that they use these compounds as carbon sources for growth.

Keywords: Polycyclic aromatic hydrocarbons (PAHs); Soil fungi; Biodegradation; Ligninolytic enzymes; Microaerobic condition; Bioremediation

Carmen Sánchez^a. (^aResearch Centre for Biological Sciences, Universidad Autónoma de Tlaxcala, Tlaxcala, México). *Lignocellulosic residues: Biodegradation and bioconversion by fungi. *Biotechnology Advances*, Volume 27(2) (2009): 185-194*

The ability of fungi to degrade lignocellulosic materials is due to their highly efficient enzymatic system. Fungi have two types of extracellular enzymatic systems; the hydrolytic system, which produces hydrolases that are responsible for polysaccharide degradation and a unique oxidative and extracellular ligninolytic system, which degrades lignin and opens phenyl rings. Lignocellulosic residues from wood, grass, agricultural, forestry wastes and municipal solid wastes are particularly abundant in nature and have a potential for bioconversion. Accumulation of lignocellulosic materials in large quantities in places where agricultural residues present a disposal problem results not only in deterioration of the environment but also in loss of potentially valuable material that can be used in paper manufacture, biomass fuel production, composting, human and animal feed among others. Several novel markets for lignocellulosic residues have been identified recently. The use of fungi in low cost bioremediation projects might be attractive given their lignocellulose hydrolysis enzyme machinery.

Keywords: Lignocellulosic residues; White-rot fungi; Lignocellulose bioconversion; Bioremediation

Abbreviations: LiP, Lignin peroxidases; MnP, manganese peroxidases; AAO, Aryl-alcohol oxidase; AAD, Aryl-alcohol dehydrogenases; QR, Quinone reductases.

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nonylphenol isomer in two agricultural soils. *Environmental Pollution*, Volume 157 (6) (2009) : 1904-1910

The degradation of a chiral nonylphenol isomer, 4-(1-ethyl-1,4-dimethylpentyl)phenol (NP₁₁₂), in two agricultural soils from Monheim and Dortmund, Germany has been studied. The degradation of NP₁₁₂ and the formation of a nitro-nonylphenol metabolite were determined by means of GC–MS analysis. The degradation followed bi-exponential order kinetics, with half-life of less than 5 days in both soils. The nitro-metabolite was found at different concentration levels in the two soils. The nitro-metabolite of NP₁₁₂ was more persistent than its parent compound. After 150 days about 13% of the initially applied NP₁₁₂ remained in the Monheim soil as its nitro-metabolite. Results of the E-screen assay revealed that the nitro-NP₁₁₂ has oestrogenic potency of 85% of that of NP₁₁₂. Furthermore, the results of chiral GC–MS analysis revealed that no chiral degradation of NP₁₁₂ occurred in this study.

The degradation of a chiral nonylphenol isomer in agricultural soils followed bi-exponential order kinetics resulting in a more persistent nitro-metabolite.

Keywords: Nitro-NP; Persistent metabolite; Estrogenic potency; E-screen; Chiral GC–MS

Mark J. Benotti^{1, a} and Bruce J. Brownawell^a. (^aMarine Sciences Research Center, Stony Brook University, Stony Brook, NY 11794-5000, USA). Microbial degradation of pharmaceuticals in estuarine and coastal seawater. *Environmental Pollution*, Volume 157(3) (2009): 994-1002

Microbial degradation rates were measured for 19 pharmaceuticals in estuarine and coastal surface water samples. Antipyrine, carbamazepine, cotinine, sulfamethoxazole, and trimethoprim were the most refractory (half-lives, $t_{1/2}$ = 35 to >100 days), making them excellent candidates for wastewater tracers. Nicotine, acetaminophen, and fluoxetine were labile across all treatments ($t_{1/2}$ = 0.68–11 days). Caffeine, diltiazem, and nifedipine were also and relatively labile in all but one of the treatments ($t_{1/2}$ = 3.5–13 days). Microbial degradation of caffeine was further confirmed by production ¹⁴CO₂. The fastest decay of non-refractory compounds was always observed in more sewage-affected Jamaica Bay waters. Degradation rates for the majority of these pharmaceuticals are much slower than reported rates for small biomolecules, such as glucose and amino acids. Batch sorption experiments indicate that removal of these soluble pharmaceuticals from the water column to sediments is a relatively insignificant removal process in these receiving waters.

Microbial degradation rates were measured for 19 structurally variable pharmaceuticals in wastewater-impacted estuarine and coastal seawater.

Keywords: PPCP; Biodegradation; Sorption; Estuary; Caffeine

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+49 89 3187 2561; fax: +49 89 3187 3361; e-mail: rainer.meckenstock@helmholtz-muenchen.de). **Anaerobic degradation of the aromatic hydrocarbon biphenyl by a sulfate-reducing enrichment culture. FEMS Microbiology Ecology, Volume 68 (1) (2009): 86 - 93**

The aromatic hydrocarbon biphenyl is a widely distributed environmental pollutant. Whereas the aerobic degradation of biphenyl has been extensively studied, knowledge of the anaerobic biphenyl-oxidizing bacteria and their biochemical degradation pathway is scarce. Here, we report on an enrichment culture that oxidized biphenyl completely to carbon dioxide under sulfate-reducing conditions. The biphenyl-degrading culture was dominated by two distinct bacterial species distantly affiliated with the Gram-positive genus *Desulfotomaculum*. Moreover, the enrichment culture has the ability to grow with benzene and a mixture of anthracene and phenanthrene as the sole source of carbon, but here the microbial community composition differed substantially from the biphenyl-grown culture. Biphenyl-4-carboxylic acid was identified as an intermediate in the biphenyl-degrading culture. Moreover, 4-fluorobiphenyl was converted cometabolically with biphenyl because in addition to the biphenyl-4-carboxylic acid, a compound identified as its fluorinated analog was observed. These findings are consistent with the general pattern in the anaerobic catabolism of many aromatic hydrocarbons where carboxylic acids are found to be central metabolites.

Keywords: biodegradation • *Desulfotomaculum* • PAH • BTEX • contamination

Gurdeep Rastogi¹, Geetha L. Muppidi¹, Raghu N. Gurram¹, Akash Adhikari¹, Kenneth M. Bischoff², Stephen R. Hughes², William A. Apel³, Sookie S. Bang¹, David J. Dixon¹ and Rajesh K. Sani¹. (¹Department of Chemical and Biological Engineering, South Dakota School of Mines and Technology, Rapid City, SD 57701, USA, ²Bioproducts and Biocatalysis Research Unit, National Center for Agricultural Utilization Research, US Department of Agriculture, Peoria, IL 61604, USA, ³Biological Systems Department, Idaho National Laboratory, Idaho Falls, ID 83415-2203, USA). **Isolation and characterization of cellulose-degrading bacteria from the deep subsurface of the Homestake gold mine, Lead, South Dakota, USA. Journal of Industrial Microbiology and Biotechnology, Volume 36(4) (2009): 585-598**

The present study investigated the cultivable mesophilic (37Â°C) and thermophilic (60Â°C) cellulose-degrading bacterial diversity in a weathered soil-like sample collected from the deep subsurface (1.5 km depth) of the Homestake gold mine in Lead, South Dakota, USA. Chemical characterization of the sample by X-ray fluorescence spectroscopy revealed a high amount of toxic heavy metals such as Cu, Cr, Pb, Ni, and Zn. Molecular community structures were determined by phylogenetic analysis of 16S rRNA gene sequences retrieved from enrichment cultures growing in presence of microcrystalline cellulose as the sole source of carbon. All phylotypes retrieved from enrichment cultures were affiliated to *Firmicutes*. Cellulose-degrading mesophilic and thermophilic pure cultures belonging to the genera *Brevibacillus*, *Paenibacillus*, *Bacillus*, and *Geobacillus* were isolated from enrichment cultures, and selected cultures were studied for enzyme activities. For a mesophilic isolate (DUSELG12), the optimum pH and temperature for carboxymethyl cellulase (CMCase) were 5.5 and 55Â°C, while for a thermophilic isolate (DUSELR7) they were 5.0 and 75Â°C, respectively. Furthermore, DUSELG12 retained about 40% CMCase activity after incubation at 60Â°C for 8 h. Most remarkably, thermophilic isolate, DUSELR7 retained 26% CMCase activity at 60Â°C up to a period of 300 h. Overall, the present work revealed the presence of different cellulose-degrading bacterial lineages in the unique deep subsurface environment of the mine. The results also have

strong implications for biological conversion of cellulosic agricultural and forestry wastes to commodity chemicals including sugars.

Keywords: Cellulose-degrading bacteria - DUSEL - Deep subsurface - Thermostable enzymes - Gold mine

Lyubov Yotova¹, Irene Tzibranska³, Filadia Tileva¹, G. H. Markx² and Nelly Georgieva¹. (¹Department of Biotechnology, University of Chemical Technology and Metallurgy, 1756 Sofia, Bulgaria, ²School of Engineering and Physical Sciences, Heriot-Watt University, Riccarton, Edinburgh, EH14 4AS, Scotland, UK, ³Department of Chemical Engineering, University of Chemical Technology and Metallurgy, 1756 Sofia, Bulgaria). **Kinetics of the biodegradation of phenol in wastewaters from the chemical industry by covalently immobilized *Trichosporon cutaneum* cells. Journal of Industrial Microbiology and Biotechnology, Volume 36(3) (2009): 367-372**

A simple method for the preparation of the biocatalyst with whole cells is presented, and the applicability of the technique for biodegradation of phenol in wastewater from the chemical industries using the basidiomycetes yeast *Trichosporon cutaneum* is explored. Kinetic studies of the influence of other compounds contained in wastewater as naphthalene, benzene, toluene and pyridine indicate that apart from oil fraction, which is removed, the phenol concentration is the only major factor limiting the growth of immobilized cells. Mathematical models are applied to describe the kinetic behavior of immobilized yeast cells. From the analysis of the experimental curves was shown that the obtained values for the apparent rate parameters vary depending on the substrate concentration ($\mu_{\max\text{app}}$ from 0.35 to 0.09 h⁻¹ and K_{sapp} from 0.037 to 0.4 g dm⁻³). The inhibitory effect of the phenol on the obtained yield coefficients was investigated too. It has been shown that covalent immobilization of *T. cutaneum* whole cells to plastic carrier beads is possible, and that cell viability and phenol degrading activity are maintained after the chemical modification of cell walls during the binding procedure. The results obtained indicate a possible future application of immobilized *T. cutaneum* for destroying phenol in industrial wastewaters.

Keywords: Biodegradation - Phenol - Immobilized cells - Kinetic parameters

V. P. Jayachandran^{1, 2} and A. A. M. Kunhi^{1, 3}. (¹Department of Food Microbiology, Central Food Technological Research Institute (CFTRI), Mysore, 570 013, Karnataka, India, ²Present address: Department of Plant Biotechnology, Mar Athanasios College for Advanced Studies, Thiruvalla, 689 101, Kerala, India, ³Present address: Central Food Laboratory, Department of Preventive Health, National Health Authority, Doha, Qatar). **Degradation of 3-chlorobenzoate and phenol singly and in mixture by a mixed culture of two ortho-pathway-following *Pseudomonas* strains. Journal of Industrial Microbiology and Biotechnology, Volume 36(2) (2009): 219-227**

The compatibility and efficiency of two ortho-cleavage pathway-following pseudomonads viz. the 3-chlorobenzoate (3-CBA)-degrader, *Pseudomonas aeruginosa* 3mT (3mT) and the phenol-degrader, *P. stutzeri* SPC-2 (SPC-2) in a mixed culture for the degradation of these substrates singly and simultaneously in mixtures was studied. Another phenol-degrading strain, *Pseudomonas* sp. SoPC-5 (SoPC-5) that utilizes a meta-cleavage mode also was tried in co-culture with 3mT. The former combination was found to be a better degrader of both the

substrates when present alone. But, with inoculum levels of 0.15 mg cell dry wt each of 3mT/SPC-2 or 3mT/SoPC-5 growth with 2 mM each of 3-CBA and phenol was slow with a lag of 24 h and degradation being incomplete. However, with higher inocula in the ratios 1:1, 1:2, and 2:1, i.e., 0.3 + 0.3, 0.3 + 0.6, and 0.6 + 0.3 mg cell dry wt of 3mT and SPC-2, respectively complete degradation of both the substrates occurred. Degradation of 3-CBA was complete with the release of stoichiometric amounts of chloride (Cl^-) when concentrations of phenol/3-CBA were varied as 2:2, 2:4, and 4:2 mM, i.e., even when the concentration of the more toxic co-substrate 3-CBA was higher than phenol effective simultaneous degradation occurred at the inoculums ratio of 1:1 (0.3 mg dry cell wt. of each strain). These studies clearly indicated the better suitability of ortho-cleavage-utilizing strains as partners in a mixed culture than those follow different modes.

Keywords: 3-Chlorobenzoate - Phenol - *P. aeruginosa* - *P. stutzeri* - Mixed culture - Simultaneous degradation

Hossein Nikakhtari *, Pardeep Kumar, Mehdi Nemati, Gordon A. Hill. (Department of Chemical Engineering, University of Saskatchewan, 57 Campus Drive Saskatoon, SK, S7N 5A9, Canada. *Biodegradation of diesel oil in a baffled roller bioreactor*. Correspondence to Hossein Nikakhtari, Department of Chemical Engineering, University of Saskatchewan, 57 Campus Drive, Saskatoon, SK, S7N 5A9, Canada). *Journal of Chemical Technology & Biotechnology*, Volume 84 (4) (2009): 525 – 532

BACKGROUND: Ex situ bioremediation is a feasible and economical way to remove petroleum pollutants from contaminated soil or water. A baffled roller bioreactor was shown to be effective for biodegradation of diesel oil as a model petroleum pollutant. Microorganisms enriched from an industrially contaminated soil with heavy hydrocarbons were shown to be the best inoculum source for diesel biodegradation.

RESULTS: The baffled roller bioreactor demonstrated better performance than control (roller bioreactor without baffles) or bead mill roller (control bioreactor filled partially with spherical beads) bioreactors. Biodegradation consisted of both fast and slow stages for degradation of light and heavy compounds, respectively. Among the tested temperatures ranging from 15 to 35 °C, room temperature (23 °C) was found to be the optimum temperature for biodegradation. The values of maximum specific growth rate and substrate yield (μ_{max} and Y_{XS}) for the indigenous microorganisms in the baffled roller bioreactor at room temperature were found to be $0.72 \pm 0.08 \text{ h}^{-1}$ and $(7.0 \pm 1.0) \times 10^7 \text{ cells mg}^{-1} \text{ diesel}$, respectively. Biodegradation of diesel concentrations up to 200 g L^{-1} was achieved with the highest biodegradation rate of $266 \text{ mg L}^{-1} \text{ h}^{-1}$ at the highest rotation rate of 45 rpm in the baffled roller bioreactor.

CONCLUSION: Using indigenous bacteria enriched from industrial contaminated soil at room temperature, a baffled roller bioreactor is able to biodegrade high diesel oil concentrations at high biodegradation rates.

Keywords: bioremediation • diesel • roller bioreactor • baffles • petroleum pollution

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Degradation of C.I. Acid Red 88 aqueous solution by combination of Fenton's reagent and ultrasound irradiation. Journal of Chemical Technology & Biotechnology, Volume 84(4) (2009): 578 – 583

BACKGROUND: Pollution caused by industrial wastewater has become a common problem for many countries. In particular, dye pollutions from industrial effluents disturb human health and ecological equilibrium. The discharge of highly colored synthetic dye effluents is aesthetically displeasing and can damage the receiving water body by impeding penetration of light. Azo dyes can be reduced to more hazardous intermediates on anaerobic conditions. Therefore, an effective and economic treatment of effluents containing a diversity of textile dyes has become a necessity for clean production technology for textile industries. Herein we wish to report the degradation of Acid Red 88 by the combination of Fenton's reagent and ultrasound irradiation.

RESULTS: The results show that the combination of ultrasonic irradiation and Fenton's reagent is effective for the degradation of Acid Red 88 aqueous solution. Furthermore, it can achieve better results than either Fenton's reagent or ultrasound alone. The optimum conditions for the degradation of Acid Red 88 aqueous solution were 1.96 mmol L⁻¹ H₂O₂, 0.108 mmol L⁻¹ Fe²⁺, pH 3.0, and ultrasonic irradiation frequency of 40 kHz. A degradation efficiency of 98.6% was achieved within 135 min.

CONCLUSION: We have provided an efficient and convenient procedure for the degradation of Acid Red 88 aqueous solution. In the present procedure, the azo linkage of Acid Red 88 is broken and some carbonyl compounds are formed, but the complete mineralization of dye cannot be achieved.

Keywords: Fenton's reagent • ultrasound irradiation • C.I. Acid Red 88 • degradation

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BACKGROUND: Mathematical models describing the physical, chemical and biological processes that take place in bioremediation are necessary to design and optimise these technologies. This work models the effect of toluene as a gaseous cosubstrate in the degradation of phenanthrene in soil, considering the consumption of pollutants, the production of intermediate degradation compounds and mineralisation. The proposed model consists of a set of sequential reactions to convert phenanthrene to carbon dioxide and biomass with the production and consumption of phthalic acid, which is the main intermediate metabolite.

RESULTS: The considerations of the model were supported by experimental data, and it was evaluated for phenanthrene degradation kinetics with previously reported packed column reactor

experiments. The mathematical model proposed describes the mineralisation of phenanthrene accurately and also predicts a reduced accumulation of phthalic acid when toluene is added as cosubstrate. The model fits the experimental data of phenanthrene degradation when toluene is added but slightly overestimates the residual phenanthrene in the control case.

CONCLUSION: The simplified model of sequential reactions represents the column experiments ($P < 0.05$) for phenanthrene degradation and mineralisation with toluene as cosubstrate, considering the production and consumption of phthalic acid.

Keywords: mathematical model • metabolites • mineralisation • phenanthrene • polycyclic aromatic hydrocarbons

Jeong Myeong Kim¹ and Che Ok Jeon¹ (¹Department of Life Science, Chung-Ang University, 221, HeukSeok-Dong, Seoul, 156-756, South Korea). **Isolation and Characterization of a New Benzene, Toluene, and Ethylbenzene Degrading Bacterium, *Acinetobacter* sp. B113. *Current Microbiology*, Volume 58(1) (2009): 70-75**

A bacterium designated strain B113, able to degrade benzene, toluene, and ethylbenzene compounds (BTE), was isolated from gasoline-contaminated sediment at a gas station in Geoje, Korea. Phylogenetic analysis based on 16S rRNA gene sequences showed that the isolate belonged to the genus *Acinetobacter*. The biodegradation rates of benzene, toluene, and ethylbenzene were relatively low in MSB broth, but the addition of yeast extract had a substantial impact on the biodegradation of BTE compounds, which suggested that yeast extract might provide a factor that was necessary for its growth or BTE biodegradation activity. However, interestingly, the biodegradation of BTE compounds occurred very quickly in slurry systems amended with sterile soil. Moreover, if soil was combusted first to remove organic matters, the enhancement effect on BTE biodegradation was lost, indicating that some insoluble organic compounds were probably beneficial for BTE degradation in contaminated sediment. This study suggests that strain B113 may play an important role for biodegradation of BTE in the contaminated site.

Baisuo Zhao¹, Hui Wang¹, Xinwei Mao¹ and Ruirui Li¹. (¹Department of Environmental Science and Engineering, Tsinghua University, Beijing, 100084, China). **Biodegradation of Phenanthrene by a Halophilic Bacterial Consortium Under Aerobic Conditions. *Current Microbiology*, Volume 58(3) (2009): 205-210**

A halophilic bacterial consortium that degraded phenanthrene was developed from oil-contaminated saline soil containing 10% salinity. The biodegradation of phenanthrene occurred at 5%, 10%, and 15% salinity, whereas no biodegradation took place at 0.1% and 20% salinity. A 16S rRNA gene analysis showed that all sequences from the denaturing gradient gel electrophoresis profile were similar to those of halophilic bacteria. This is the first report of a halophilic bacterial consortium capable of degrading phenanthrene under hypersaline conditions.

Qing Hong^{a, b}, Xiaojun Dong^b, Lijuan He^b, Xin Jiang^a and Shunpeng Li^b. (^aState Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Science, Nanjing, 210008, China, ^bKey Laboratory of Microbiological Engineering Agricultural Environment, Ministry of Agriculture, College of Life Science, Nanjing Agricultural University, Nanjing 210095, China). **Isolation of a biphenyl-degrading**

bacterium, *Achromobacter* sp. BP3, and cloning of the *bph* gene cluster. International Biodeterioration & Biodegradation, Volume 63(4) (2009): 365-370

A bacterial strain, BP3, capable of degrading biphenyl, was isolated from petroleum-contaminated soil. Strain BP3 was identified preliminarily as *Achromobacter* sp. based on its physiological and biochemical characteristics and 16S rRNA gene sequence analysis. Strain BP3 was able to degrade 50 mg l⁻¹ of biphenyl within 12 h. A 16.7-kb DNA fragment consisting of the entire *bph* cluster (*bphRA1A2XA3A4BCKHJID*) was obtained by normal PCR amplification and chromosome walking. Genes encoding integrase and transposon related genes were detected upstream and downstream of the *bph* cluster, respectively, which indicated that the *bph* cluster might locate on a big mobile genetic element (MGE).

Keywords: Biphenyl; Degradation; *bph* cluster; Mobile genetic element

Hui Wang^{a, b}, Jian Qiang Su^{a, b}, Xiao Wei Zheng^{a, b}, Yun Tian^a, Xiao Jing Xiong^b and Tian Ling Zheng^{a, b}. (^aKey Laboratory of Ministry of Education for Coast and Wetland Ecosystems, School of Life Sciences, Xiamen University, Xiamen 361005, China, ^bState Key Laboratory of Marine Environmental Science, College of Marine Science & Environmental Science, Xiamen University, Xiamen 361005, China). **Bacterial decolorization and degradation of the reactive dye Reactive Red 180 by *Citrobacter* sp. CK3^{*} International Biodeterioration & Biodegradation, Volume 63(4) (2009): 395-399**

A bacterial strain, CK3, with remarkable ability to decolorize the reactive textile dye Reactive Red 180, was isolated from the activated sludge collected from a textile mill. Phenotypic characterization and phylogenetic analysis of the 16S rDNA sequence indicated that the bacterial strain belonged to the genus *Citrobacter*. Bacterial isolate CK3 showed a strong ability to decolorize various reactive textile dyes, including both azo and anthraquinone dyes. Anaerobic conditions with 4 g l⁻¹ glucose, pH = 7.0 and 32 °C were considered to be the optimum decolorizing conditions. *Citrobacter* sp. CK3 grew well in a high concentration of dye (200 mg l⁻¹), resulting in approximately 95% decolorization extent in 36 h, and could tolerate up to 1000 mg l⁻¹ of dye. UV-vis analyses and colorless bacterial cells suggested that *Citrobacter* sp. CK3 exhibited decolorizing activity through biodegradation, rather than inactive surface adsorption. It is the first time that a bacterial strain of *Citrobacter* sp. has been reported with decolorizing ability against both azo and anthraquinone dyes. High decolorization extent and facile conditions show the potential for this bacterial strain to be used in the biological treatment of dyeing mill effluents.

Keywords: *Citrobacter* sp. CK3; Decolorization; Degradation; Reactive Red 180

Esmaeil AL-Saleh^a, Hana Drobiova^a and Christian Obuekwe^a. (^aMicrobiology Program, Department of Biological Sciences, College of Science, Kuwait University, P.O. Box 5969, Safat 13060, Kuwait). **Predominant culturable crude oil-degrading bacteria in the coast of Kuwait. International Biodeterioration & Biodegradation, Volume 63(4) (2009): 400-406**

Total of 272 crude oil-degrading bacteria were isolated from seven locations along the coast of Kuwait. The analysis of the 16S rDNA sequences of isolated bacteria revealed the predominance of six bacterial genera: *Pseudomonas*, *Bacillus*, *Staphylococcus*, *Acinetobacter*, *Kocuria* and

Micrococcus. Investigation of the factors associated with bacterial predominance revealed that, dominant culturable crude oil-degrading bacteria were better crude oil utilizers than the less frequently occurring isolates. Bacterial predominance was also influenced by the ability of bacteria to adapt to the level of organic content available. Predominant culturable bacteria constituted 89.7–54.2% of the total crude oil-degrading bacterial communities. Using 16S-RFLP analyses to assess the diversity of the dominant crude oil-degrading bacterial genera, four phylotypes of *Pseudomonas* sp. and seven phylotypes of *Bacillus* sp. were determined. This suggested high degree of diversity of crude oil-degrading bacterial population at the strain level, but low diversity at the genus level.

Keywords: Crude oil; Biodegradation; 16S rDNA; Phylogeny; Coast; Diversity

Nedra Asses^a, Lamia Ayed^a, Hassib Bouallagui^a, Sami Sayadi^b and Moktar Hamdi^a. (^aLaboratoire d'Ecologie et de Technologie Microbienne, Institut National des Sciences Appliquées et de Technologie (INSAT), 2 Boulevard de la terre, B.P. 676, 1080 Tunis, Tunisie, ^bLaboratoire des Bioprocédés, Centre de Biotechnologie de Sfax, BP: «K» 3038 Sfax, Tunisie). **Biodegradation of different molecular-mass polyphenols derived from olive mill wastewaters by *Geotrichum candidum*. International Biodeterioration & Biodegradation, Volume 63(4) (2009): 407-413**

The decolourisation of fresh and stored olive mill wastewaters (OMW) and the biodegradation of three groups (F1, F2 and F3) of phenolic compounds by *Geotrichum candidum* were investigated. Separated phenolic compounds derived from natural OMW ultrafiltration using membranes with a cutoff 2 and 100 kDa. *G. candidum* growth on fresh OMW decreased pH and reduced COD and colour of 75% and 65%, respectively. However, on the stored-black OMW a failure of COD and colour removal were observed. *G. candidum* activity on this later substrate was enhanced by the addition of a carbon source easily metabolised, misleading an improvement of the COD reduction and decolourization that reached 58% and 48%, respectively. Growth of *G. candidum* in the presence of F2 or F3 polyphenolic fractions induced high decolourisation and depolymerisation of phenolic compounds. Whereas, very weak decolourisation and biodegradation were observed with F1 fraction. Moreover, the highest levels of lignin peroxidase (LiP) and manganese peroxidase (MnP) activities were obtained in the presence of F2 fraction. These results showed that increasing of molecular-mass of aromatics led to an increase in levels of depolymerisation, decolourisation and COD removal by *G. candidum* culture.

Keywords: *Geotrichum candidum*; Olive mill wastewater; Polyphenols; Decolourisation; Lignin peroxidase; Manganese peroxidase

Gajanan Ghodake^a, Sheetal Jadhav^b, Vishal Dawkar^a and Sanjay Govindwar^a. (^aDepartment of Biochemistry, Shivaji University, Vidyanagar, Kolhapur 416 004, Maharashtra, India, ^bDepartment of Microbiology, Shivaji University, Vidyanagar, Kolhapur 416 004, Maharashtra, India). **Biodegradation of diazo dye Direct brown MR by *Acinetobacter calcoaceticus* NCIM 2890. International Biodeterioration & Biodegradation, Volume 63(4) (2009): 433-439**

Acinetobacter calcoaceticus was employed for the degradation of Direct brown MR (DBMR), commercially used azo dye in the textile industry in order to analyze mechanism of the degradation and role of inhibitors, redox mediators and stabilizers of lignin peroxidase during decolorization. Induction of intracellular and extracellular lignin peroxidase, intracellular laccase

and DCIP reductase represented their involvement in the biodegradation of DBMR. Decolorization and biodegradation of azo dye DBMR in broth were monitored by UV-visible spectrophotometer and TLC. The products obtained from *A. calcoaceticus* degradation were characterized by FTIR and identified by GC/MS as biphenyl amine, biphenyl, 3-amino 6-hydroxybenzoic acid and naphthalene diazonium. Germination (%) and growth efficiency of *Sorghum vulgare* and *Phaseolus mungo* seeds revealed the degradation of DBMR into less toxic products than original dye. *A. calcoaceticus* also has a potential to degrade diverse dyes present in the textile effluent, into nontoxic metabolites, hence *A. calcoaceticus* can be applied for the commercial application.

Keywords: *A. calcoaceticus*; Biodegradation; DBMR; Lignin peroxidase; *Sorghum vulgare*

Katarína Dercová^a, Jana Šeligová^a, Hana Dudášová^a, Mária Mikulášová^b, Katarína Šilhárová^c, Lívia Tóthová^c and Pavel Hucko^c. (^aSlovak University of Technology, Faculty of Chemical and Food Technology, Institute of Biotechnology and Food Science, Department of Biochemical Technology, Radlinského 9, 812 37 Bratislava, Slovakia, ^bInstitute of Cell Biology, Faculty of Natural Sciences, Mlynská dolina, Comenius University, 842 15 Bratislava, Slovakia, ^cWater Research Institute, Nábřežie arm. gen. L. Svobodu 5, 812 49 Bratislava, Slovakia). **Characterization of the bottom sediments contaminated with polychlorinated biphenyls: Evaluation of ecotoxicity and biodegradability. International Biodeterioration & Biodegradation, Volume 63(4) (2009): 440-449**

At the locality of the former producer of PCBs Chemko Strážske in East Slovakia, a large amount of PCBs (the commercial mixture DELOR 103, an equivalent of AROCLOR 1242) is still persisting in sediments and negatively influences health of the population. The objective of this work was to provide a study of ecotoxicity and genotoxicity of PCBs in contaminated sediments. Toxicity of the PCB-contaminated sediments sampled from Zemplínska šírava and Strážsky canal (surroundings of the former producer of PCBs) was determined applying a standard aquatic plant toxicity test using *Lemna minor*. The endpoints for the test were frond numbers and frond areas. The sediment sampled from Zemplínska šírava was more toxic to *L. minor* than the one sampled from Strážsky canal. The results on genotoxicity showed that both sediments were not mutagenic toward the standard strains of the Ames test, *Salmonella typhimurium* TA98 and TA100. This work deals also with biodegradation of PCBs in two samples of the above mentioned contaminated sediments: a) in the natural sediments by autochthonous microbial consortium and b) in the bioaugmented sediments inoculated by allochthonous bacterial strains, two bacterial isolates from long-term PCB-contaminated soil *Pseudomonas stutzeri* and *Alcaligenes xylosoxidans*. Both approaches were applied under the biostimulation conditions, with addition of glucose or biphenyl as co-substrates, as well. The highest PCB degradation was observed in the bioaugmented sediment inoculated with bacterial strain *P. stutzeri*. Addition of biphenyl, as the co-substrate and the inducer, positively affected degradation of PCBs. The *bphA1* gene, encoding enzyme biphenyldioxygenase, responsible for the start of PCB degradation, was identified in genome of *P. stutzeri*, a potential PCB-degrader isolated from long-term PCB-contaminated soil, but not in genome of *A. xylosoxidans*.

Keywords: Biodegradation; *bphA1* gene; Ecotoxicity; *Pseudomonas stutzeri*; Polychlorinated biphenyls; Sediments

Ji-Guang Gu^a, Boping Han^a, Shunshan Duan^a, Zhenye Zhao^b and Yuping Wang^b. (^aDepartment of Ecology, and Institute of Hydrobiology, Jinan University, Guangzhou 10015, PR China, ^bLaboratory of Environmental Toxicology, School of Biological Sciences, The University of Hong Kong, Pokfulam Road, Hong Kong SAR, PR China). **Degradation of the endocrine-disrupting dimethyl phthalate carboxylic ester by *Sphingomonas yanoikuyae* DOS01 isolated from the South China Sea and the biochemical pathway. *International Biodeterioration & Biodegradation*, Volume 63(4) (2009): 450-455**

Bacteria capable of using dimethyl phthalate (DMP) as the sole carbon and energy source were isolated from the sediments collected at a depth of 1340 m from the South China Sea. *Sphingomonas yanoikuyae* DOS01, identified based on 16S rRNA gene sequence, utilized DMP from an initial level of 180 mg l⁻¹ to non-detectable in 35 h at 30 °C, the optical density (OD₆₀₀) values increased over the time of incubation. Degradation intermediate monomethyl phthalate (MMP) accumulated up to 21.3 mg l⁻¹ and then disappeared in the culture medium. When MMP or another intermediate phthalate (PA) was used as the sole substrate, this strain was only capable of degrading MMP, but not PA. Total organic carbon (TOC) analysis of the culture medium suggested that both DMP and MMP were mineralized, but not PA. This strain from the deep-ocean sediment transforms DMP to MMP using a common biochemical pathway for DMP as reported before. Further esterase activity assays indicated that the enzyme induced by MMP has higher affinity than that by DMP for the substrate *p*-nitrophenyl acetate. Our results indicated that complete degradation of DMP by this marine microorganism may involve a new biochemical pathway.

Keywords: Degradation; Biochemical pathways; Deep-ocean; Dimethyl phthalate; Endocrine-disruption

Mads Pedersen^a, Morten Hollensted^a, Lene Lange^b and Birgitte Andersen^a. (^aDepartment of Systems Biology, Søtofts Plads, Building 221, Technical University of Denmark, DK-2800 Lyngby, Denmark, ^bNovozymes A/S, Smørumsevej, DK-2880 Bagsværd, Denmark). **Screening for cellulose and hemicellulose degrading enzymes from the fungal genus *Ulocladium*. *International Biodeterioration & Biodegradation*, Volume 63(4) (2009): 484-489**

The fungal genus *Ulocladium* consists mostly of saprotrophic species and can readily be isolated from dead vegetation, rotten wood, paper, textiles and other cellulose containing materials. Thus, they must produce cellulolytic and hemicellulolytic enzymes. In this study fifty *Ulocladium* strains from ten different species were tested for enzyme activities on 14 different azurine-cross-linked (AZCL) substrates and analyzed by multivariate analysis. The tested strains of *Ulocladium* were found to produce a broad enzyme profile. Most species in *Ulocladium* were able to produce high amounts of enzymes that degraded amylose, arabinoxylan, β-glucan, cellulose and xylan; however, variations between species as well as between individual strains in each species were seen. Overall, the enzyme profiles were found to be species specific, but also source of isolation impacted the enzymes produced. The results suggest that species identity as well as isolation source must be considered when screening microorganisms for enzymes.

Keywords: Azurine-cross-linked; AZCL; Enzyme production; Wheat bran agar; Species specificity; Source specificity

Tejomye S. Bhalerao^a and Pravin R. Puranik^a. (^aSchool of Life Sciences, North Maharashtra University, P.B. No. 80, Jalgaon, Maharashtra 425001, India). **Microbial degradation of monocrotophos by *Aspergillus oryzae*. International Biodeterioration & Biodegradation, Volume 63(4) (2009): 503-508**

Organophosphorus pesticides are widely used in India for protection of agricultural yields. However, these pesticides pose various threats to organisms, including humans, and hamper soil microbial activity; thus, they are a cause for concern. As a measure of bioremediation, soil fungi capable of degrading monocrotophos (MCP) were isolated from various geographical and ecological sites. Twenty-five strains were isolated by an enrichment method using MCP as a carbon and phosphorus source. On the basis of MCP tolerance capacity exhibited in gradient agar plate assay the isolate M-4, identified as *Aspergillus oryzae* ARIFCC 1054, was selected for further studies. The ability of the isolate to mineralize MCP was investigated under different culture conditions. The isolate was found to possess phosphatase activity. The course of the degradation process was studied using HPTLC and FTIR analyses. The results suggest that this organism could be used for bioaugmentation of soil contaminated with MCP and for treatment of aqueous wastes.

Keywords: *Aspergillus oryzae*; MCP; Monocrotophos; Organophosphorus pesticide

R.J. Varma^a and B.G. Gaikwad^a. (^aChemical Engineering Division, National Chemical Laboratory, Pashan Road, Pune 411008, India). **Biodegradation and phenol tolerance by recycled cells of *Candida tropicalis* NCIM 3556. International Biodeterioration & Biodegradation, Volume 63(4) (2009): 539-542**

Resting cells of *Candida tropicalis* NCIM 3556 rapidly degraded almost completely 2 g L⁻¹ phenol in 16 h. In this study, we explored the possibility of further increasing the efficiency of the culture by repeatedly reusing the cell for biodegradation. The effect of continuous recycling of whole cells of *C. tropicalis*, for biodegradation of phenol indicated that though with each recycle of the cell there was steady decline in phenol biodegradation the conversion was appreciable for five recycle (~70%) and reached half-life of 50% after eleven recycles. Inhibition due to substrate, recycling of cells and adaptation of residual cell were estimated and an equation derived; which indicated that the cell resilience to phenol increased with each cycle and at the end of eleven recycle adaptation was 68%. However, when the adapted cells were sub cultured and showed marginal increase <10% in biodegradation.

Keywords: *Candida tropicalis*; Phenol; Tolerance; Biodegradation; Recycle; Inhibition

Yii Siang Hii^a, Ah Theem Law^a, N.A.M. Shazili^a, M.K. Abdul-Rashid^a and Choon Weng Lee^b. (^aInstitute of Oceanography, University Malaysia Terengganu, 21030 Kuala Terengganu, Terengganu, Malaysia, ^bInstitute of Biological Science, University Malaya, 50603 Kuala Lumpur, Malaysia). **Biodegradation of *Tapis* blended crude oil in marine sediment by a consortium of symbiotic bacteria. International Biodeterioration & Biodegradation, Volume 63(2) (2009): 142-150**

Biodegradation rate and the high molecular weight hydrocarbons are among the important concerns for bioremediation of crude oil. Inoculation of a non-oil-degrading bacterium as

supplementary bacteria increased oil biodegradation from 57.1% to 63.0% after 10 days of incubation. Both the oil-degrading bacteria and the non-oil-degrading bacteria were isolated from Malaysian marine environment. Based on the 16S rDNA sequences, the oil-degrading bacteria was identified as *Pseudomonas pseudoalcaligenes* (99% similarity) while the non-oil-degrading bacterium was *Erythrobacter citreus* (99% similarity). *E. citreus* does not grow on crude oil enriched medium under present experimental condition but it withstands 5000 mg kg⁻¹ *Tapis* blended crude oil in sediment. Under optimal condition, the oil-degrading bacterium; *P. pseudoalcaligenes*, alone utilized 583.3 ± 3.8 mg kg⁻¹ (57.1%) at the rate of 3.97 × 10⁻¹⁰ mg kg⁻¹ cell⁻¹ day⁻¹ *Tapis* blended crude oil from 1000 mg kg⁻¹ oil-contaminated sediment. Inoculation of *E. citreus* as the supplementary bacteria to *P. pseudoalcaligenes* enhanced biodegradation. The bacterial consortium degraded 675.8 ± 18.5 mg kg⁻¹ (63.0%) *Tapis* blended crude oil from the 1000 mg kg⁻¹ oil-contaminated sediment. Biodegradation rate of the bacterial consortium increased significantly to 4.59 × 10⁻¹⁰ mg kg⁻¹ cell⁻¹ day⁻¹ (*p* = 0.02). Improvement of the oil degradation by the bacterial consortium was due to the synergetic reaction among the bacterial inoculants. There are two implications: (1) *E. citreus* may have a role in removing self-growth-inhibiting compounds of *P. pseudoalcaligenes*. (2) *P. pseudoalcaligenes* degraded *Tapis* blended crude oil while *E. citreus* competes for the partially degraded hydrocarbons by *P. pseudoalcaligenes*. *P. pseudoalcaligenes* forced to breakdown more hydrocarbons to sustain its metabolic requirement. The bacterial consortium degraded 78.7% of (C₁₂–C₃₄) total aliphatic hydrocarbons (TAHs) and 74.1% of the 16 USEPA prioritized polycyclic aromatic hydrocarbons.

Keywords: *Tapis* blended crude oil; Biodegradation; Sediment; Bacterial consortium; Symbiosis

Luis Diorio^a, Beatriz Galati^a, María Amela García^a and Leandro Papinutti^a. (^aDepartment of Biodiversity and Experimental Biology, Faculty of Exact and Natural Sciences, University of Buenos Aires, Argentina). **Degradation of pruning wastes by *Phanerochaete sordida* growing in SSF: Ultrastructural, chemical, and enzymatic studies. International Biodeterioration & Biodegradation, Volume 63(1) (2009): 19-23**

A solid standard fermentation (SSF) with the fungus *Phanerochaete sordida* in a medium with *Nephrolepis cordifolia* (entire pinnae separated from the rachis) and *Laurus nobilis* (fragmented leaves) was performed over 92 days to study the degradation of leaves with histological, chemical, and enzymatic methods. The fungus entered the leaves early, through the stomata in *N. cordifolia* and *L. nobilis*, and also through mechanical cuts that had been made in the latter. The initial attack affected the mesophyll in both plant species, and the phloem in *L. nobilis*. The vascular bundle of *N. cordifolia* was protected by a sheath of cells with thick lignified walls. The collenchyma cell walls situated near the principal vein in *L. nobilis* swelled during the initial stages of enzymatic action, but reduced their thickness afterwards, mainly in regions of contact with the hyphae. At the end of the experiment, no species had leaves with mesophyll. In *L. nobilis*, phloem was also lacking, and a partial and heterogeneous attack on the xylem became evident. The histological changes are compared with the enzymatic activities and the chemical composition of the culture media, describing the stages of fungal colonization.

Keywords: *Phanerochaete sordida*; Pruning wastes; Biodeterioration; Ultrastructure; Fungal exoenzymes

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600036, India, ^bPolymer Research and Technology center, Reliance Industries Limited, V.N. PuravMarg, Mumbai-600071, India). Degradation of untreated and thermally pretreated polypropylene by soil consortia. International Biodeterioration & Biodegradation, Volume 63(1) (2009): 106-111

Untreated (PP-UT) and thermally pretreated (at 80 °C for 10 days) polypropylene (PP-TT) films of 0.05 mm thickness were subjected to *in vitro* biodegradation in minimal medium with mixed soil culture for 12 months. In this period 10.7 and 0.4% weight loss was observed with PP-TT and PP-UT, respectively. The tensile strength decreased by 51.8 and 28.3%, the crystallinity increased by 28 and 33% and isotacticity increased by 3 and 9%, respectively, over the same time period. The ester carbonyl index in PP-TT increased up to 9 months and later decreased indicating abiotic followed by biotic process. No such changes were observed with PP-UT. Methyl group index decreased in both the cases indicating oxidation at the primary carbon. Increase in surface energy indicated that the polymer became hydrophilic. Surface changes were observed by SEM and AFM. A single culture was isolated at the end of 12 months and it was identified as *Bacillus flexus*. The morphology of the organism was rods in a chain and it was present in the form of an endospore.

Keywords: *Bacillus flexus*; Thermal pretreatment; Biodegradation; FTIR

Andreas Stolz¹. (¹Institut für Mikrobiologie der Universität Stuttgart, Allmandring 31, 70569 Stuttgart, Germany). Molecular characteristics of xenobiotic-degrading sphingomonads. Applied Microbiology and Biotechnology, Volume 81(5) (2009): 793-811

The genus *Sphingomonas* (*sensu lato*) belongs to the α -Proteobacteria and comprises strictly aerobic chemoheterotrophic bacteria that are widespread in various aquatic and terrestrial environments. The members of this genus are often isolated and studied because of their ability to degrade recalcitrant natural and anthropogenic compounds, such as (substituted) biphenyl(s) and naphthalene(s), fluorene, (substituted) phenanthrene(s), pyrene, (chlorinated) diphenylether(s), (chlorinated) furan(s), (chlorinated) dibenzo-*p*-dioxin(s), carbazole, estradiol, polyethylene glycols, chlorinated phenols, nonylphenols, and different herbicides and pesticides. The metabolic versatility of these organisms suggests that they have evolved mechanisms to adapt quicker and/or more efficiently to the degradation of novel compounds in the environment than members of other bacterial genera. Comparative analyses demonstrate that sphingomonads generally use similar degradative pathways as other groups of microorganisms but deviate from competing microorganisms by the existence of multiple hydroxylating oxygenases and the conservation of specific gene clusters. Furthermore, there is increasing evidence for the existence of plasmids that only can be disseminated among sphingomonads and which undergo after conjugative transfer pronounced rearrangements.

Keywords: *Sphingomonas* - Sphingomonads - Biodegradation - Xenobiotics - Degradative plasmids

Daisuke Sugimori¹. (¹Department of Industrial System, Faculty of Symbiotic Systems Science, Fukushima University, 1 Kanayagawa, Fukushima 960-1296, Japan). Edible oil degradation by using yeast coculture of *Rhodotorula pacifica* ST3411 and *Cryptococcus laurentii* ST3412. Volume 82(2) (2009): 351-357

To develop a microbial treatment of edible oil-contaminated wastewater, microorganisms capable of rapidly degrading edible oil were screened. The screening study yielded a yeast coculture comprising *Rhodotorula pacifica* strain ST3411 and *Cryptococcus laurentii* strain ST3412. The coculture was able to degrade efficiently even at low contents of nitrogen ($[\text{NH}_4\text{-N}] = 240 \text{ mg/L}$) and phosphorus sources ($[\text{PO}_4\text{-P}] = 90 \text{ mg/L}$). The 24-h degradation rate of 3,000 ppm mixed oils (salad oil/lard/beef tallow, 1:1 w/w) at 20°C was $39.8\% \pm 9.9\%$ (means \pm standard deviations of eight replicates). The highest degradation rate was observed at 20°C and pH 8. In a scaled-up experiment, the salad oil was rapidly degraded by the coculture from 671 ± 52.0 to 143 ± 96.7 ppm in 24 h, and the degradation rate was $79.4\% \pm 13.8\%$ (means \pm standard deviations of three replicates). In addition, a repetitive degradation was observed with the cell growth by only pH adjustment without addition of the cells.

Keywords: Oil degradation - Coculture - *Rhodotorula* - *Cryptococcus* - Wastewater treatment

Olusola A. Ojo^{1*} and Benjamin A. Oso². (¹Department of Microbiology, Lagos State University, Badagry Expressway, P.O. Box 12142, Ikeja, Lagos-Nigeria, ²Department of Botany/Microbiology, University of Ibadan, Nigeria. *Corresponding author. E-mail:solayom@yahoo.com. Tel: +234 – 8055055478). **Biodegradation of synthetic detergents in wastewater. African Journal of Biotechnology Volume 8 (6) (2009): 1090–1109**

A total of 76 wastewater samples were randomly collected from pharmaceutical, textile, and detergent-manufacturing industries as well as the Agbara Sewage Treatment Plant (STP). Thirty-eight samples each in 2-L plastic containers were collected for morning and evening effluent used for this study. Composite samples were later developed and the physico-chemical properties of these samples determined. The physico-chemical properties of the composite wastewater influenced the selected microbial population adapted to utilization of detergent components. The optimum temperature range of the composite wastewater was $33.9 - 34.3^\circ\text{C}$ while the mean optimum pH ranged from 6.9 – 8.8 for the laboratory simulated biodegradation of test detergents. Although, the fungal consortium was eliminated as the medium approached the alkaline pH, this is as a result of the metabolites produced. The macro-elements, the BOD and the hydrocarbon concentration of the composite effluent were above the EU and FEPA limits for discharged effluent. The composite effluent was thereafter spiked with test detergents (Elephant, Omo, Klin, Ariel Persil, Teepol, and SDS) at 0.01% (w/v) and its progressive degradation monitored for 30 days. The microbial detergent-degraders population changed between Day 0 and 15, thereafter it stabilized. The heterotrophic bacterial count from the seventy-six randomly collected effluent samples was 42.9×10^6 cfu/ml, while the mean bacterial detergent-degrader population was 20.94×10^6 cfu/ml. The mean fungal population from the randomly collected effluent sample was 4.5×10^6 cfu/ml. The bacterial detergent-degraders characterized and identified include *Pseudomonas aeruginosa*, *Escherichia coli*, *Enterococcus majodoratus*, *Klebsiella liquefasciens*, *Enterobacter liquefasciens*, *Klebsiella aerogenes*, *Enterobacter agglomerans*, *Staphylococcus albus*, *Proteus* sp., *Klebsiella oxytoca* and *Brevibacterium* sp., while the fungal detergent-degrader included; *Myceliophthora thermophila*, *Geomyces* sp., *Alternaria alternata*, *Fusarium* sp., *Aspergillus flavus* and *Asperigillus oryzae*. The primary biodegradability of synthetic detergent was confirmed by the Methylene Blue–Active Substance (MBAS) method. Gas chromatography (GC) provided the convincing evidence

of synthetic detergent mineralization within the 30 day period in a sewage treatment plant. The detection of unusual peaks in the GC profiles provided the scientific evidence of inclusion of certain hydrocarbons in detergent formulation outside that of industry specifications. The unusual peaks are attributable to inclusion of certain chemical optical brighteners (C₁₇-C₂₄). Linear alkyl benzene sulphonates (LAS) which is the principal synthetic detergent component are thus biodegradable and its use in detergent formulation is environment - friendly.

Keywords: Biodegradation, detergents, linear alkylbenzene sulphonate, sustainable development.

Ania C. Ulrich¹, Selma E. Guigard¹, Julia M. Foght², Kathleen M. Semple², Kathryn Pooley¹, James E. Armstrong³ and Kevin W. Biggar^{1,4}. (¹Department of Civil and Environmental Engineering, University of Alberta, Edmonton, AB, Canada, T6G 2W2, ²Department of Biological Sciences, University of Alberta, Edmonton, AB, Canada, ³WorleyParsons Komex International Inc., Calgary, AB, Canada, ⁴Present address: BGC Engineering Inc., Edmonton, AB, Canada). **Effect of salt on aerobic biodegradation of petroleum hydrocarbons in contaminated groundwater. Biodegradation, Volume 20(1) (2009): 27-38**

Hydrocarbon-contaminated soil and groundwater at oil and gas production sites may be additionally impacted by salts due to release of produced waters. However, little is known about the effect of salt on the in-situ biodegradation of hydrocarbons by terrestrial microbes, especially at low temperatures. To study this effect, we prepared a groundwater-soil slurry from two sites in Canada: a former flare pit site contaminated with flare pit residue (Site A), and a natural gas processing facility contaminated with natural gas condensate (Site B). The slurry with its indigenous microbes was amended with radiolabeled hydrocarbons dissolved in free product plus nutrients and/or NaCl, and incubated in aerobic biometer flasks with gyrotory shaking at either 25 or 10°C for up to 5 weeks. Cumulative production of ¹⁴CO₂ was measured and the lag time, rate and extent of mineralization were calculated. For Site A, concentrations of NaCl ≥1% (w/v) delayed the onset of mineralization of both ¹⁴C-hexadecane and ¹⁴C-phenanthrene under nutrient-amended conditions, but once biodegradation began the degradation rates were similar over the range of salt concentrations tested (0–5% NaCl). For Site B, increasing concentrations of NaCl ≥1% (w/v) increased the lag time and decreased the rate and extent of mineralization of aliphatic and aromatic substrates. Of particular interest is the observation that low concentrations of salt (≤1% NaCl) slightly stimulated mineralization in some cases.

Keywords: Aerobic biodegradation - Groundwater - Petroleum hydrocarbons - Pollutants - Salt

Yousuke Suzuki¹ and Noriyuki Koyama¹. (¹Department of Chemistry, Faculty of Science, Chiba University, Yayoi Chiba, 263-8522, Japan). **Uptake and degradation of EDTA by Escherichia coli. Biodegradation, Volume 20(1) (2009): 39-44**

It was found that *Escherichia coli* exhibited a growth by utilization of Fe(III)EDTA as a sole nitrogen source. No significant growth was detected when Fe(III)EDTA was replaced by EDTA complexes with other metal ions such as Ca²⁺, Co²⁺, Cu²⁺, Mg²⁺, Mn²⁺, and Zn²⁺. When EDTA uptake was measured in the presence of various ions, it was remarkable only when Fe³⁺ was present. The cell extract of *E. coli* exhibited a significant degradation of EDTA only in the

presence of Fe³⁺. It is likely that the capability of *E. coli* for the growth by utilization of Fe(III)EDTA results from the Fe³⁺-dependent uptake and degradation of EDTA.

Keywords: EDTA uptake - EDTA degradation - Fe³⁺-dependent

P. N. Tallur¹, V. B. Megadi¹ and H. Z. Ninnekar¹. (¹Department of Biochemistry, Karnataka University, Dharwad, 580003, Karnataka, India). **Biodegradation of p -cresol by immobilized cells of Bacillus sp. strain PHN 1. Biodegradation, Volume 20 (1) (2009): 79-83**

The *Bacillus* sp. strain PHN 1 capable of degrading *p*-cresol was immobilized in various matrices namely, polyurethane foam (PUF), polyacrylamide, alginate and agar. The degradation rates of 20 and 40 mM *p*-cresol by the freely suspended cells and immobilized cells in batches and semi-continuous with shaken cultures were compared. The PUF-immobilized cells achieved higher degradation of 20 and 40 mM *p*-cresol than freely suspended cells and the cells immobilized in polyacrylamide, alginate and agar. The PUF-immobilized cells could be reused for more than 35 cycles, without losing any degradation capacity and showed more tolerance to pH and temperature changes than free cells. These results revealed that the immobilized cell systems are more efficient than freely suspended cells for degradation of *p*-cresol.

Keywords: Degradation - Immobilization - *p*-Cresol - Polyurethane foam - *Bacillus* sp. strain PHN 1

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The 2-ethylhexyl nitrate (2-EHN) is currently added to diesel oil to improve ignition and boost cetane number. The biodegradability of this widely used chemical needed to be assessed in order to evaluate the environmental impact in case of accidental release. In aerobic liquid cultures, biodegradation of 2-EHN was assessed in biphasic liquid cultures using an inert non-aqueous phase liquid such as 2,2,4,4,6,8,8-heptamethylnonane (HMN) as solvent for the hydrophobic substrate. 2-EHN was found to be biodegradable by microbial communities from refinery wastewater treatment plants, but was recalcitrant to those of urban wastewater treatment facilities. Out of eighteen hydrocarbon-polluted or non-polluted soil samples, six microbial populations were also able to degrade 2-EHN. However, strain isolation from these microbial populations was rather difficult suggesting close cooperation between members of the microbial communities. Specific axenic bacterial strains selected for their ability to catabolize recalcitrant-hydrocarbons were also tested for their capacity to degrade 2-EHN. In liquid cultures with HMN phase as non-aqueous phase liquid, some *Mycobacterium austroafricanum* strains were found to degrade and mineralize 2-EHN significantly.

Keywords: 2-Ethylhexyl nitrate - Biodegradation - Mineralization - Soil microbial population - *Mycobacterium*

Janice C. Paslawski¹, John V. Headley², Gordon A. Hill³ and Mehdi Nemati³. (¹Division of Environmental Engineering, University of Saskatchewan, 57 Campus Drive, Saskatoon, Saskatchewan, Canada, S7N 5A9, ²Water Science and Technology Directorate, 11 Innovation Boulevard, Saskatoon, Saskatchewan, Canada, S7N 3H5, ³Department of Chemical Engineering, University of Saskatchewan, 57 Campus Drive, Saskatoon, Saskatchewan, Canada, S7N 5A9). **Biodegradation kinetics of trans-4-methyl-1-cyclohexane carboxylic acid. Biodegradation, Volume 20(1) (2009): 125-133**

Naphthenic acids are a complex mixture of organic compounds which naturally occur in crude oil. Low molecular weight components of the naphthenic acids are known to be toxic in aquatic environments and there is a need to better understand the factors controlling the kinetics of their biodegradation. In this study, a relatively low molecular weight naphthenic acid compound (trans-isomer of 4-methyl-1-cyclohexane carboxylic acid) and a microbial culture developed in our laboratory were used to study the biodegradation of this naphthenic acid and to evaluate the kinetics of the process in batch cultures. The initial concentration of trans-4-methyl-1-cyclohexane carboxylic acid (50–750 mg l⁻¹) did not affect the maximum specific growth rate of the bacteria at 23°C (0.52 day⁻¹) to the maximum biodegradable concentration (750 mg l⁻¹). The maximum yield observed at this temperature and at a neutral pH was 0.21 mg of biomass per milligram of substrate. Batch experiments indicated that biodegradation can be achieved at low temperatures; however, the biodegradation rate at room temperature (23°C) and neutral pH was 5 times faster than that observed at 4°C. Biodegradation at various pH conditions indicated a maximum specific growth rate of 1.69 day⁻¹ and yield (0.41 mg mg⁻¹) at a pH of 10.

Keywords: Biodegradation - Kinetics - Naphthenic acids

Supriya Goswami¹ and Dileep K. Singh¹. (¹Department of Zoology, University of Delhi, Delhi, 110007, India). **Biodegradation of $\hat{I}\pm$ and \hat{I}^2 endosulfan in broth medium and soil microcosm by bacterial strain *Bordetella* sp. B9. Biodegradation, Volume 20(2) (2009):199-207**

Bacterial strains were isolated from endosulfan treated soil to study the microbial degradation of this pesticide in broth medium and soil microcosm. The isolates were grown in minimal medium and screened for endosulfan degradation. The strain, which utilized endosulfan and showed maximum growth, was selected for detail studies. Maximum degrading capability in shake flask culture was shown by *Bordetella* sp. B9 which degraded 80% of α endosulfan and 86% of β endosulfan in 18 days. Soil microcosm study was also carried out using this strain in six different treatments. Endosulfan ether and endosulfan lactone were the main metabolites in broth culture, while in soil microcosm endosulfan sulfate was also found along with endosulfan ether and endosulfan lactone. This bacterial strain has a potential to be used for bioremediation of the contaminated sites.

Keywords: *Bordetella* sp. B9 - Biodegradation - Soil microcosm

Muftah H. El-Naas^a, Shaheen A. Al-Muhtaseb^{1, a} and Souzan Makhlof^a. (^aChemical & Petroleum Engineering Department, U.A.E. University, P.O. Box 17555, Al-Ain, United Arab Emirates). **Biodegradation of phenol by *Pseudomonas putida* immobilized in**

polyvinyl alcohol (PVA) gel. Journal of Hazardous Materials, Volume 164(2-3) (2009): 720-725

Batch experiments were carried out to evaluate the biodegradation of phenol by *Pseudomonas putida* immobilized in polyvinyl alcohol (PVA) gel pellets in a bubble column bioreactor at different conditions. The bacteria were activated and gradually acclimatized to high concentrations of phenol of up to 300 mg/l. The experimental results indicated that the biodegradation capabilities of *P. putida* are highly affected by temperature, pH, initial phenol concentration and the abundance of the biomass. The biodegradation rate is optimized at 30 °C, a pH of 7 and phenol concentration of 75 mg/l. Higher phenol concentrations inhibited the biomass and reduced the biodegradation rate. At high phenol concentration, the PVA particle size was found to have negligible effect on the biodegradation rate. However, for low concentrations, the biodegradation rate increased slightly with decreasing particle size. Other contaminants such heavy metals and sulfates showed no effect on the biodegradation process. Modeling of the biodegradation of phenol indicated that the Haldane inhibitory model gave better fit of the experimental data than the Monod model, which ignores the inhibitory effects of phenol.

Keywords: Biodegradation; Immobilization; Phenol; Wastewater; Refinery

Huili Wang^a, Shuxia Xu^b, Chengxia Tan^c and Xuedong Wang^d. (^aCollege of Life Sciences, Wenzhou Medical College, Wenzhou 325035, China, ^bDepartment of Microbiology, College of Life Sciences, Henan Agricultural University, Zhengzhou 450002, China, ^cCollege of Chemical Engineering and Material Science, Zhejiang University of Technology, Hangzhou 310014, China, ^dSchool of Environmental Science and Public Health, Wenzhou Medical College, Wenzhou 325035, China). **Anaerobic biodegradation of hexazinone in four sediments. Journal of Hazardous Materials, Volume 164(2-3) (2009): 806-811**

Anaerobic biodegradation of hexazinone was investigated in four sediments (L1, L2, Y1 and Y2). Results showed that the L2 sediment had the highest biodegradation potential among four sediments. However, the Y1 and Y2 sediments had no capacity to biodegrade hexazinone. Sediments with rich total organic carbon, long-term contamination history by hexazinone and neutral pH may have a high biodegradation potential because the former two factors can induce the growth of microorganisms responsible for biodegradation and the third factor can offer suitable conditions for biodegradation. The addition of sulfate or nitrate as electron acceptors enhanced hexazinone degradation. As expected, the addition of electron donors (lactate, acetate or pyruvate) substantially inhibited the degradation. In natural environmental conditions, the effect of intermediate A [3-(4-hydroxycyclohexyl)-6-(dimethylamino)-1-methyl-1,3,5-triazine-2,4(1H, 3H)dione] on anaerobic hexazinone degradation was negligible because of its low level.

Keywords: Hexazinone; Anaerobic (anoxic); Sediment; Electron donor; Electron acceptor

Tony Hadibarata^a, Sanro Tachibana^a and Kazutaka Itoh^a. (^aDepartment of Applied Bioscience, Faculty of Agriculture, Ehime University, 3-5-7 Tarumi, Matsuyama, Ehime 790-8566, Japan). **Biodegradation of chrysene, an aromatic hydrocarbon by *Polyporus* sp. S133 in liquid medium. Journal of Hazardous Materials, Volume 164(2-3) (2009): 911-917**

Polyporus sp. S133, a fungus collected from contaminated-soil was used to degrade chrysene, a polycyclic aromatic hydrocarbon (PAH) in a mineral salt broth (MSB) liquid culture. Maximal

degradation rate of chrysene (65%) was obtained when *Polyporus* sp. S133 was incubated in the cultures supplemented with polypeptone (10%) for 30 days under agitation of 120 rpm, as compared to just 24% degradation rate in non-agitated culture. Furthermore, the degradation of chrysene was affected by the addition of carbon and nitrogen sources as well as kind of surfactants. The degradation rate was increased with increase in added amount of carbon and nitrogen sources, respectively. The degradation rate in agitated cultures was enhanced about 2 times higher than that in non-agitated cultures. The degradation mechanism of chrysene by *Polyporus* sp. S133 was determined through identification of several metabolites; chrysenequinone, 1-hydroxy-2-naphthoic acid, phthalic acid, salicylic acid, protocatechuic acid, gentisic acid, and catechol. Several enzymes (manganese peroxidase, lignin peroxidase, laccase, 1,2-dioxygenase and 2,3-dioxygenase) produced by *Polyporus* sp. S133 were detected during the incubation. The highest enzyme activity was shown by 1,2-dioxygenase (237.5 U l^{-1}) after 20 days of incubation.

Keywords: Chrysene; Biodegradation; Metabolites; *Polyporus* sp. S133

Jingshun Zhang^{b, c}, Zhongtao Sun^b, Yingying Li^{b, c}, Xiang Peng^{b, c}, Wen Li^{b, c} and Yanchun Yan^a. (^aGraduate School, Chinese Academy of Agricultural Sciences, Beijing 100081, PR China, ^bCollege of Life Sciences, Shandong Agricultural University, Tai'an, Shandong 271018, PR China, ^cState Key Laboratory of Crop Biology, Shandong Agricultural University, Tai'an, Shandong 271018, PR China). **Biodegradation of *p*-nitrophenol by *Rhodococcus* sp. CN6 with high cell surface hydrophobicity. *Journal of Hazardous Materials*, Volume 163(2-3) (2009): 723-728**

Rhodococcus sp. CN6, isolated from a pesticide industry's effluent-sediment, was able to completely degrade and utilize 100 mg/L *p*-nitrophenol (PNP) as the sole carbon, nitrogen and energy sources for growth in the minimal salt media (MSM) within 12 h. To study the applicability of the strain for bioremediation of PNP, its degradation potential was examined in the presence of different supplemented carbon and nitrogen sources in MSM with 100 mg/L PNP. Dextrin was experienced as the best supplemented carbon source used by the strain CN6 during degrading PNP. Addition of ammonium nitrate could also increase the PNP degradation rate. Preliminary studies on the surface characters of *Rhodococcus* sp. CN6 were undertaken for the sake of exploring its high efficiency on the degradation of PNP. Microbial adherence to hydrocarbons (MATH) assays illuminated that the strain CN6 was of higher hydrophobicity while grown on higher concentration of PNP. The results suggested that the strain CN6 could be used as a potential and efficient PNP degrader for the bioremediation of contaminated sites.

Keywords: *p*-Nitrophenol; Degradation; *Rhodococcus* sp. CN6; Hydrophobicity

J. Paca^a, M. Halecky^a, J. Barta^a and R. Bajpai^b. (^aDepartment of Fermentation Chemistry and Bioengineering, Institute of Chemical Technology, Prague, Technicka 5, 166 28, Czech Republic, ^bDepartment of Chemical Engineering, University of Louisiana, Lafayette, LA 70504, USA). **Aerobic biodegradation of 2,4-DNT and 2,6-DNT: Performance characteristics and biofilm composition changes in continuous packed-bed bioreactors. *Journal of Hazardous Materials*, Volume 163(2-3) (2009): 848-854**

This manuscript deals with continuous experiments for biodegradation of individual dinitrotoluenes by a defined mixed culture in packed-bed reactors (PBRs) containing either poraver or fire-clay as packing material. Removal efficiencies and volumetric biodegradation rates were measured as a function of the loading rate of 2,4-dinitrotoluene (2,4-DNT) and 2,6-dinitrotoluene (2,6-DNT) under steady-state conditions. The poraver reactor showed higher removal efficiencies for both the DNTs. The removal efficiency for 2,4-DNT remained greater than 90% in the poraver reactor whereas it dropped steadily from 85 to 65% in the fire-clay reactor as the organic loading rates were increased from 19 to 60 mg L⁻¹ day⁻¹. Similar trends were seen for the volumetric degradation rate as well. In both the reactors, 2,4-DNT degraded more effectively than 2,6-DNT. The microbial consortium was characterized both in the inoculum as well as in the operating PBR. Cell numbers per gram dry packing material were similar in the two reactors. However, there was a distinct difference in the nature of microorganisms that were found in the two packings. The fire-clay contained a larger number of cells that were not primary degraders of DNTs.

Keywords: Dinitrotoluenes; Biodegradation; Packed-bed reactor; Biofilm composition; Degradation efficiency

Abbreviations: BSM, basal salt medium; CFM, colony forming microorganisms; DNT, dinitrotoluene; DOC, dissolved oxygen concentration; HRT, hydraulic retention time (day); OL, organic loading (mg L⁻¹ day⁻¹); PBR, packed-bed reactor; RE, removal efficiency (%); SEM, scanning electron microscopy; STDEV, standard deviation; TNT, trinitrotoluene

Jinyou Shen^a, Jianfa Zhang^a, Yi Zuo^a, Lianjun Wang^a, Xiuyun Sun^a, Jiansheng Li^a, Weiqing Han^a and Rui He^a. (^aSchool of Chemical Engineering, Nanjing University of Science and Technology, Nanjing 210094, Jiangsu Province, China). **Biodegradation of 2,4,6-trinitrophenol by *Rhodococcus* sp. isolated from a picric acid-contaminated soil. Journal of Hazardous Materials, Volume 163(2-3) (2009): 1199-1206**

A picric acid-degrading bacterium, strain NJUST16, was isolated from a soil contaminated by picric acid and identified as a member of *Rhodococcus* sp. based on 16S rRNA sequence. The degradation assays suggested that the strain NJUST16 could utilize picric acid as the sole source of carbon, nitrogen and energy. The isolate grew optimally at 30 °C and initial pH 7.0–7.5 in the mineral salts medium supplemented with picric acid. It was basically consistent with degradation of picric acid by the isolate. Addition of nitrogen sources such as yeast extract and peptone accelerated the degradation of picric acid. However, the stimulation was concentration dependent. The degradation was accompanied by release of stoichiometric amount of nitrite and acidification. The degradation of picric acid at relatively high concentrations (>3.93 mM) demonstrated that the degradation was both pH and nitrite dependent. Neutral and slightly basic pH was crucial to achieve high concentrations of picric acid degradation by the NJUST16 strain.

Keywords: Picric acid; *Rhodococcus* sp. NJUST16; Biodegradation; Nitroaromatic compounds

Fatma Gurbuz^a, Hasan Ciftci^b and Ata Akcil^b. (^aSuleyman Demirel University, Department of Biological Sciences, TR32260 Isparta, Turkey, ^bSuleyman Demirel University, Department of Mining Engineering, Mineral Processing Division, BIOMIN Group, TR32260 Isparta, Turkey). **Biodegradation of cyanide containing effluents by *Scenedesmus obliquus*. Journal of Hazardous Materials, Volume 162(1) (2009): 74-79**

Biological degradation of cyanide has been shown a viable and robust process for degrading cyanide in mining process wastewaters. Several algal cultures can effectively degrade cyanide as carbon and/or nitrogen source for their growth. In this study, cyanide effluent degradation by *Scenedesmus obliquus* was examined. Gold mill effluents containing WAD cyanide concentration of 77.9 mg/L was fed to batch unit to examine the ability of *S. obliquus* for degrading cyanide. Cyanide was reduced down to 6 mg/L in 77 h. Microbial growth and metal uptake of Zn, Fe and Cu was examined during cyanide degradation. The cells well adapted to high pH and the effluent contained cyanide and the metals. It is important that Zn level reduced down 50%, of the starting concentration. pH was kept at 10.3 to prevent loss of cyanide as HCN, due its volatile nature. The bio treatment process was considered to be successful in degrading cyanide in the mine process water.

Keywords: Cyanide; Degradation; Gold mining; *Scenedesmus obliquus*; Environment

M.P. Elizalde-González^a, L.E. Fuentes-Ramírez^b and M.R.G. Guevara-Villa^{a, b}. (^aCentro de Química, Instituto de Ciencias, Universidad Autónoma de Puebla, Apdo. Postal J-55, Puebla, Pue. 72571, Mexico, ^bCentro de Investigación en Ciencias Microbiológicas, Instituto de Ciencias, Universidad Autónoma de Puebla, Apdo. Postal J-55, Puebla, Pue. 72571, Mexico). **Degradation of immobilized azo dyes by *Klebsiella* sp. UAP-b5 isolated from maize bioadsorbent. *Journal of Hazardous Materials*, Volume 161(2-3) (2009): 769-774**

The degradation of two immobilized dyes by *Klebsiella* sp. UAP-b5 was studied. In batch experiments, the azo dyestuffs Basic Blue 41 and Reactive Black 5 were immobilized onto corn cobs by adsorption, and the adsorption process was characterized by a pseudo-second-order kinetic equation. *Klebsiella* sp. UAP-b5 was previously isolated from the corn waste and shown to decolorize these dyes in liquid systems. Here, we demonstrate anaerobic decolorization and reductive biodegradation of these dyes by means of spectrophotometry, HPLC, and IR spectroscopy of the solid waste and desorption solutions. We also demonstrate adsorption of compounds that resemble known degradation products.

Keywords: Reactive black 5; Biodegradation; *Klebsiella*; Maize waste; Immobilization

Biosensor

M.K. Gilmanov^a, A.R. Kerimkulova^a, A.N. Sabitov^a and S.A. Ibragimova^a. (^aLaboratory of structure and regulation of enzymes, M.A. Aytkhozhin's Institute of molecular biology and biochemistry, Almaty, Kazakhstan). **The phosphatidylinositol–protein nanocomplex as a new biosensor for ecological monitoring and clinical diagnostic. *Biosensors and Bioelectronics*, Volume 24(5) (2009): 1490-1492**

Using chromatography on nanostructured carbon sorbent we had isolated an unusual nanocomplex from filling grains of wheat and maize, which consists of only phosphatidylinositol (PI) and one protein—glutamate dehydrogenase (GDh). It was very surprising that this nanocomplex shows activity of Nicotinamide adenine dinucleotide phosphate-GDh (NADP-GDh) without any treatment. Thus, the whole body of nanocomplex shows its activity without disturbing its integrity. This makes the nanocomplex very convenient for using it as a biosensor. The main feature of nanocomplex is its high sensitivity to ammonia ions. Linear response

concentration for nanocomplex is from 0.5 μM to 10 μM ammonia ions. Due to these properties the nanocomplex may be very useful as nanobiosensor for ecological monitoring of pollution by sewer waters of natural reservoirs—lakes and rivers. Also this nanosensor can be applied for determination of ammonia ions, NADPH and 2-oxoglutarate in biological liquids for clinical diagnostic.

Keywords: Glutamate dehydrogenase; Nanocomplex; Biosensor; Ecological monitoring

Javier García-Alonso^a, Gillian M. Greenway^a, Joerg D. Hardege^a and Stephen J. Haswell^a. (^aFaculty of Science, The University of Hull, Hull HU6 7RX, England, United Kingdom). **A prototype microfluidic chip using fluorescent yeast for detection of toxic compounds. Biosensors and Bioelectronics, Volume 24(5) (2009): 1508-1511**

A microfluidic chip has been developed to enable the screening of chemicals for environmental toxicity. The microfluidic approach offers several advantages over macro-scale systems for toxicity screening, including low cost and flexibility in design. This design flexibility means the chips can be produced with multiple channels or chambers which can be used to screen for different toxic compounds, or the same toxicant at different concentrations. *Saccharomyces cerevisiae* containing fluorescent markers are ideal candidates for the microfluidic screening system as fluorescence is emitted without the need of additional reagents. Microfluidic chips containing eight multi-parallel channels have been developed to retain yeast within the chip and allow exposure of them to toxic compounds. The recombinant yeast used was GreenScreen™ which expresses green fluorescent proteins when is exposed to genotoxins. After exposure of the yeast to target compounds, the fluorescence emission was detected using an inverted microscope. Qualitative and quantitative comparisons of the fluorescent emission were performed. Results indicated that fluorescent intensity per area significantly increases upon exposure to methyl-methanesulfonate, a well known genotoxic compound.

The microfluidic approach reported here is an excellent tool for cell-based screening and detection of different toxicities. The device has the potential for use by industrial manufacturers to detect and reduce the production and discharge of toxic compounds, as well as to characterise already polluted environments.

Keywords: Microfluidic chip; GFP; Toxicity screening; Yeast

Hsieh-Cheng Han^a, Ying-Rong Chang^a, Wen-Lin Hsu^{b, 1} and Chien-Yuan Chen^{a, b, 1}. (^aDepartment of Biochemical Science and Technology and Institute of Microbiology and Biochemistry, National Taiwan University, Taipei, Taiwan, ROC, ^bDepartment of Radiation Oncology, Buddhist Tzu Chi General Hospital, Taichung Branch, Taichung, Taiwan, ROC). **Application of parylene-coated quartz crystal microbalance for on-line real-time detection of microbial populations. Biosensors and Bioelectronics, Volume 24(6) (2009): 1543-1549**

A novel technique of applying a quartz crystal microbalance (QCM) sensor to the on-line real-time detection of microbial populations is described. The *p*QCM sensor was fabricated by depositing di-para-xylene (parylene) over the entire surface of a QCM sensor through a chemical vapor deposition (CVD) process. An electrically insulated film of parylene on the QCM sensor enabled the operation of the sensor in the liquid environment, and the resonance frequency of the

*p*QCM sensor set in the medium of a cultivation flask shifted in response to the microbial population.

The effects of pH, conductivity, and viscosity of the medium on the frequency shift of the *p*QCM sensor were investigated. Ignorable responses (less than 1% at 10^3 cells) were obtained during an incubation cycle.

The detection limit of the *p*QCM sensor was identified as 10^2 cells ml^{-1} with a frequency shift of around 2×10^3 Hz. The cell numbers of *Escherichia coli* cultivated in both the YEM medium and whole milk were detected. A satisfactory correlation ($r^2 = 0.95$) was obtained between the cell number and the response of the *p*QCM sensor.

Experimental results suggest that the *p*QCM described here is applicable to the continuous long-term detection of microbial populations during a fermentation process.

Keywords: Quartz crystal microbalance; Parylene; *p*QCM sensor; *E. coli*; Mass loading; Fermentation process

L. De Stefano^a, L. Rotiroti^a, M. De Stefano^b, A. Lamberti^c, S. Lettieri^d, A. Setaro^d and P. Maddalena^d. (^aUnit of Naples-Institute for Microelectronics and Microsystems, Via P. Castellino 111, 80131 Naples, Italy, ^bDepartment of Environmental Sciences, Second University of Naples, Via Vivaldi 43, 81100 Caserta, Italy, ^cDepartment of Biochemistry and Medical Biotechnologies, University of Naples “Federico II”, Via Pansini 5, 80100 Naples, Italy, ^dCNR-INFM Coherentia and Department of Physical Sciences, University of Naples “Federico II”, Via Cinthia, 80126 Naples, Italy). **Marine diatoms as optical biosensors. *Biosensors and Bioelectronics*, Volume 24(6) (2009): 1580-1584**

We have chemically modified the frustules of the marine diatom *Coscinodiscus concinnus* Wm. Smith to properly bind a highly selective bioprobe such as an antibody. By measuring the changes in the photoluminescence emission of diatoms frustules, we have monitored the molecular recognition event between the antibody and its ligand: the dissociation constant estimated is of the same order of that measured by standard Biacore[®]. The nanostructured silica frustules, a low-cost and natural available material, have shown high sensitivity, equal to $1.2 \pm 0.2 \text{ nm } \mu\text{M}^{-1}$, and a detection limit of 100 nM, and thus are quite ideal candidates for lab-on-particle applications.

Keywords: Optical biosensors; Diatoms; Photoluminescence

Javier Ramón-Azcón^a, Ryouta Kunikata^b, F.-J. Sanchez^a, M.-P. Marco^a, Hitoshi Shiku^b, Tomoyuki Yasukawa^b and Tomokazu Matsue^b. (^aApplied Molecular Receptors Group (AMRg), IQAB-CSIC, 18-26 Jordi Girona, Barcelona 08034, Spain, ^bGraduate School of Environmental Studies, Tohoku University, 6-6-11 Aoba, Aramaki, Aoba, Sendai 980-8579, Japan). **Detection of pesticide residues using an immunodevice based on negative dielectrophoresis. *Biosensors and Bioelectronics*, Volume 24(6) (2009): 1592-1597**

The detection of atrazine using a novel optical immunosensing technique based on negative dielectrophoresis (n-DEP) in microfluidic channels is described. Atrazine is a toxic triazine

herbicide within the most frequently used. Polystyrene microparticles (6 μm diameters) modified with bovine serum albumin conjugated with atrazine (atrazine-BSA) were manipulated and captured when subjected to intense n-DEP electric fields. Specifically, particles were trapped when AC voltages with amplitudes of 10 V_{peak} and frequencies over 1 MHz were applied to the electrodes. The immunological reaction occurring on the particles for detecting atrazine is based on an indirect competitive assay using a secondary anti-mouse immunoglobulin G (IgG) antibody labeled with fluorescein isothiocyanate. The microfluidic device, with three-dimensional microelectrodes, was fabricated comprising two caged areas, allowing two simultaneous measurements inside the same microfluidic channel. The performance of this n-DEP immunosensing technique was evaluated using wine samples. The immunodevice showed a limit of detection for atrazine in buffer samples of $0.11 \mu\text{g L}^{-1}$ and in pre-treated wine samples of $6.8 \mu\text{g L}^{-1}$; these detection limits are lower than the maximum residue level (MRL) established by the European Community for residues of this herbicide in wine ($50 \mu\text{g L}^{-1}$). This methodology offers great promise for rapid, simple, cost effective, and on-site analysis of biological, foods and beverages, and environmental samples.

Keywords: Immunodevice; Antibodies; Dielectrophoresis; Microfluidic device; Microparticles; Atrazine; Wine matrix effect

Sung-Rok Hong^a, Suk-Jung Choi^a, Hyun Do Jeong^b and Suhee Hong^a. (^aFaculty of Marine Bioscience and Technology, Department of Chemistry, Kangnung National University, Kangnung 210-702, Republic of Korea, ^bDepartment of Aquatic Life Medicine, Pukyong National University, Busan 608-737, Republic of Korea). **Development of QCM biosensor to detect a marine derived pathogenic bacteria *Edwardsiella tarda* using a novel immobilisation method. Biosensors and Bioelectronics, Volume 24(6) (2009) : 1635-1640**

QCM technology offers a real time output, simplicity of use and cost effectiveness in addition to high sensitivity. Sensitivity of QCM immunosensor can be enhanced by improving the immobilisation procedure on the quartz surface. The immobilisation strategy should be able to control both the amount and the orientation of the antibody (immunoglobulin; IgG) on the transducer for high affinity to antigens. This study introduced a new methodology recruiting oxidised IgG to expose aldehyde group in Fc region to cross-link to hydrazide conformed on self assembled monolayer (SAM) and compared with three conventional methods. Consequently, it was proved that considerable amount of antibody was immobilised and the sensitivity of new methodology was higher than other methods while ability of new methodology to immobilise IgG was lower than the conventional methods. The frequency shifts following bacterial cell injection were positively related to the frequency shifts after the injection of IgG and the amounts of bacterial cells, revealing that the frequency shifts after bacterial cell injection fully represented the weight change by specific attachments of bacterial cells to the IgG cross-linked on the gold surface. Specificity was tested on different bacteria including *E. coli*, *V. vulnificus* and *A. hydrophila* and showed no significant non-specific affinity on the tested bacteria. It was also demonstrated that the prepared sensor chip was stable enough to withstand repeated surface regeneration. Indeed, polyclonal antibody was more effective to detect antigen than monoclonal antibody which binds to only one epitope of antigen. Conclusively, the new methodology is appeared to be more sensitive than conventional methods tested and reusable for 10 times.

Keywords: QCM; Immunosensor; Immobilisation; Regeneration; *Edwardsiella tarda*

Padmapriya P. Banada^a, Karleigh Huff^a, Euiwon Bae^c, Bartek Rajwa^d, Amornrat Aroonual^a, Bulent Bayraktar^e, Abrar Adil^a, J. Paul Robinson^{d, f}, E. Daniel Hirleman^c and Arun K. Bhunia^a. (^aMolecular Food Microbiology Laboratory, Department of Food Science, Purdue University, IN 47907, USA, ^cSchool of Mechanical Engineering, Purdue University, USA, ^dDepartment of Basic Medical Sciences, Purdue University, USA, ^eSchool of Electrical and Computer Engineering, Purdue University, USA, ^fWeldon School of Biomedical Engineering, Purdue University, USA). **Label-free detection of multiple bacterial pathogens using light-scattering sensor. Biosensors and Bioelectronics, Volume 24(6) (2009): 1685-1692**

Technologies for rapid detection and classification of bacterial pathogens are crucial for securing the food supply. This report describes a light-scattering sensor capable of real-time detection and identification of colonies of multiple pathogens without the need for a labeling reagent or biochemical processing. Bacterial colonies consisting of the progeny of a single parent cell scatter light at 635 nm to produce unique forward-scatter signatures. Zernike moment invariants and Haralick descriptors aid in feature extraction and construction of the scatter-signature image library. The method is able to distinguish bacterial cultures at the genus and species level for *Listeria*, *Staphylococcus*, *Salmonella*, *Vibrio*, and *Escherichia* with an accuracy of 90–99% for samples derived from food or experimentally infected animal. Varied amounts of exopolysaccharide produced by the bacteria causes changes in phase modulation distributions, resulting in strikingly different scatter signatures. With the aid of a robust database the method can potentially detect and identify any bacteria colony essentially instantaneously. Unlike other methods, it does not destroy the sample, but leaves it intact for other confirmatory testing, if needed, for forensic or outbreak investigations.

Keywords: Light scatterometer; Bacterial detection; Classification; Food; Clinical specimen

Angel Valero-Navarro^a, Alfonso Salinas-Castillo^b, Jorge F. Fernández-Sánchez^a, Antonio Segura-Carretero^a, Ricardo Mallavia^b and Alberto Fernández-Gutiérrez^a. (^aDepartment of Analytical Chemistry, University of Granada, c/Fuentenueva s/n, 18071 Granada, Spain¹, ^bInstitute of Molecular and Cellular Biology, University Miguel Hernandez, Elche, Spain). **The development of a MIP-optosensor for the detection of monoamine naphthalenes in drinking water. Biosensors and Bioelectronics, Volume 24(7) (2009): 2305-2311**

To enhance the advantages of fluorescent flow-through sensing for drinking water we have designed a novel sensing matrix based on molecularly imprinted polymers (MIPs). The synergic combination of a tailor-made MIP recognition with a selective room temperature fluorescence detection is a novel concept for optosensing devices and is assessed here for the simple and selective determination of pollutants in water.

We describe a simple approach to preparing synthetic receptors for monoamine naphthalene compounds (MA-NCs) using non-covalent molecular imprinting techniques and naphthalene as template. We examine in detail the binding characteristics of the imprinted polymer and describe the flow-through sensor of MA-NCs by solid-surface fluorescence. Its detection limits for recognizing 1-naphthylamine (1-NA) and 2-naphthylamine (2-NA) separately are 26 ng mL⁻¹ and 50 ng mL⁻¹, respectively, and it also determines 1-NA and 2-NA simultaneously with a detection limit of 45 ng mL⁻¹.

All the instrumental, chemical and flow variables were carefully optimized and an interference study was carried out to demonstrate its applicability and selectivity. Finally, we applied it to the analysis of 1-NA and 2-NA in tap and mineral waters, obtaining a 98% average recovery rate.

Keywords: Molecular imprinting; Fluorescence optosensor; Flow injection; Naphthalene compounds; Water analysis

Ri Mi Lee^a, Hyangtae Choi^b, Jeon-Soo Shin^c, Kunhong Kim^b, and Kyung-Hwa Yoo^a. (^aNanomaterial Graduate Program and Department of Physics, Yonsei University, 134 Shinchon-dong, Seodaemun-gu, Seoul 120-749, Republic of Korea, ^bDepartment of Biochemistry and Molecular Biology and Center for Chronic Metabolic Disease Research, Yonsei University College of Medicine, Brain Korea 21 Project for Medical Science of Yonsei University, 134 Shinchon-dong, Seodaemun-gu, Seoul 120-752, Republic of Korea, ^cDepartment of Microbiology, Yonsei University College of Medicine, Brain Korea 21 Project for Medical Science of Yonsei University, 134 Shinchon-dong, Seodaemun-gu, Seoul 120-752, Republic of Korea). **Distinguishing between apoptosis and necrosis using a capacitance sensor. Biosensors and Bioelectronics, Volume 24(8) (2009): 2586-2591**

Apoptosis and necrosis are two different paths for cell death. One of differences between apoptosis and necrosis is the cell morphology. Apoptotic cells shrink without losing the integrity of their plasma membrane and break into smaller pieces called apoptotic bodies that other body cells recognize and eat. In contrast, necrotic cells swell and their plasma membrane eventually ruptures. Since the cell membrane is closely related to the capacitance (or dielectric constant), we have fabricated a capacitance sensor, which can measure the capacitance of cells, and investigated its time dependence during apoptosis and necrosis for TE2 cells induced by TNF-related apoptosis inducing ligand (TRAIL) and ethanol. The capacitance decreases monotonically during apoptosis. For necrosis, however, step-like behaviors are observed and dips are found in the $dC/dt-t$ curves. The time-lapse images of TE2 cells, which have been taken simultaneously with the capacitance measurements, show that the dips in the $dC/dt-t$ curves are probably due to the rupture of cell membrane. These results suggest that apoptosis and necrosis are differentiated by the capacitance measurements.

Keywords: Capacitance sensor; Cell proliferation; Apoptosis; Necrosis

Shili Liu^a, Kenny K. Tran^a, Steven Pan^a and Hong Shen^a. (^aDepartment of Chemical Engineering, University of Washington, Box 351750, Seattle, WA 98195, United States). **Detecting and differentiating microbes by dendritic cells for the development of cell-based biosensors. Biosensors and Bioelectronics, Volume 24(8) (2009): 2598-2603**

Dendritic cells (DCs) are a specialized family of antigen presenting cells. They play critical roles in sensing and processing microbial information through a series of pattern recognition receptors (PPRs), including the well-characterized toll-like receptors (TLRs). In this study, we demonstrated the utilization of a DC cell line, DC2.4, as a cell source for the detection and differentiation of microbes towards the development of cell-based biosensors. As a proof of principle, the Gram-negative bacteria *Escherichia coli* K12 strain D21 and its lipopolysaccharide (LPS) mutants were used as model targets. The stimulation of DCs by bacterial strains was monitored by the production of nitric oxide (NO), and the colorimetric Greiss assay was used to quantify the level of NO produced. Our results demonstrated that DCs could detect and differentiate microbes with subtle differences in the composition of specific cell surface

components, i.e. LPS, within minutes. Though the current colorimetric-based NO assay limited the detection sensitivity, we showed that DCs were able to detect as low as 2–3 bacteria per cell. Furthermore, compared to macrophages, DCs were superior in discriminating LPS mutants. Our study demonstrates that DCs possess great potential as cell sources for the development of novel cell-based biosensors for detecting microbes with high selectivity and sensitivity and rapid responsiveness. In addition, when DCs are coupled with other biosensor platforms, higher sensitivity can be expected.


Keywords: Immune cells; Lipopolysaccharide; Nitric oxide (NO); Toll-like receptors

Bioengineering

Xueming Tang,^{1,2*} Yongsong Tan,¹ Hong Zhu,¹ Kai Zhao,¹ and Wei Shen² (Biotechnology Research Institute, Shanghai Academy of Agricultural Sciences, 2901 Beidi Road, Shanghai 201106, People's Republic of China,¹ School of Biotechnology and Key Lab for Industrial Biotechnology of Education Ministry, Southern Yangtze University, 1800 Lihudadao Avenue, Wuxi, Jiangsu 214122, People's Republic of China²). Microbial Conversion of Glycerol to 1,3-Propanediol by an Engineered Strain of *Escherichia coli*. Applied and Environmental Microbiology, Volume 75 (6) (2009): 1628-1634

In an effort to improve industrial production of 1,3-propanediol (1,3-PD), we engineered a novel polycistronic operon under the control of the temperature-sensitive lambda phage P_LP_R promoter regulated by the *cIts857* repressor and expressed it in *Escherichia coli* K-12 ER2925. The genes for the production of 1,3-PD in *Clostridium butyricum*, *dhaB1* and *dhaB2*, which encode the vitamin B₁₂-independent glycerol dehydratase DhaB1 and its activating factor, DhaB2, respectively, were tandemly arrayed with the *E. coli yqhD* gene, which encodes the 1,3-propanediol oxidoreductase isoenzyme YqhD, an NADP-dependent dehydrogenase that can directly convert glycerol to 1,3-PD. The microbial conversion of 1,3-PD from glycerol by this recombinant *E. coli* strain was studied in a two-stage fermentation process. During the first stage, a novel high-cell-density fermentation step, there was significant cell growth and the majority of the metabolites produced were organic acids, mainly acetate. During the second stage, glycerol from the fresh medium was rapidly converted to 1,3-PD following a temperature shift from 30°C to 42°C. The by-products were mainly pyruvate and acetate. During this two-stage process, the overall 1,3-PD yield and productivity reached 104.4 g/liter and 2.61 g/liter/h, respectively, and the conversion rate of glycerol to 1,3-PD reached 90.2% (g/g). To our knowledge, this is the highest reported yield and productivity efficiency of 1,3-PD with glycerol as the sole source of carbon. Furthermore, the overall fermentation time was only 40 h, shorter than that of any other reports.

Jodi Switzer Blum,¹ Sukkyun Han,² Brian Lanoil,³ Chad Saltikov,⁴ Brian Witte,⁵ F. Robert Tabita,⁵ Sean Langley,⁶ Terry J. Beveridge,^{7,†} Linda Jahnke,⁸ and Ronald S. Oremland^{1*} (U.S. Geological Survey, Menlo Park, California 94025,¹ Department of Environmental Sciences, University of California, Riverside, California 92521,² Department of Biological Sciences, University of Alberta, Edmonton, Alberta T6G 2E1, Canada,³ Department of Microbiology and Environmental Toxicology, University of California, Santa Cruz,

California 95064,⁴ Department of Microbiology, The Ohio State University, Columbus, Ohio 43210,⁵ Department of Earth Sciences, University of Ottawa, Ottawa, Ontario K1N 6N5, Canada,⁶ Department of Molecular and Cellular Biology, University of Guelph, Guelph, Ontario N1G 2W1, Canada,⁷ NASA Ames Research Center, Moffett Field, California 94035⁸). Ecophysiology of "*Halarsenatibacter silvermanii*" Strain SLAS-1^T, gen. nov., sp. nov., a Facultative Chemoautotrophic Arsenate Respirer from Salt-Saturated Searles Lake, California . Applied and Environmental Microbiology, Volume 75 (7) (2009): 1950-1960,

Searles Lake occupies a closed basin harboring salt-saturated, alkaline brines that have exceptionally high concentrations of arsenic oxyanions. Strain SLAS-1^T was previously isolated from Searles Lake (R. S. Oremland, T. R. Kulp, J. Switzer Blum, S. E. Hoefft, S. Baesman, L. G. Miller, and J. F. Stolz, Science 308:1305-1308, 2005). We now describe this extremophile with regard to its substrate affinities, its unusual mode of motility, sequenced *arrABD* gene cluster, cell envelope lipids, and its phylogenetic alignment within the order *Halanaerobacteriales*, assigning it the name "*Halarsenatibacter silvermanii*" strain SLAS-1^T. We also report on the substrate dynamics of an anaerobic enrichment culture obtained from Searles Lake that grows under conditions of salt saturation and whose members include a novel sulfate reducer of the order *Desulfovibrionales*, the archaeon *Halorhabdus utahensis*, as well as a close homolog of strain SLAS-1^T.

C R Kokare*, S Chakraborty, A N Khopade and K R Mahadik. (Department of Pharmaceutical Biotechnology, Poona College of Pharmacy, Bharati Vidyapeeth University, Pune 411 038, India). Biofilm: Importance and applications. Indian Journal of Biotechnology, Volume 8(2009) :159-168

Biofilm is an assemblage of the microbial cells that is irreversibly associated with a surface and usually enclosed in a matrix of polysaccharide material. Biofilm is composed primarily of microbial cells and extracellular polymeric substance (EPS). Extracellular polymeric matrix plays various roles in structure and function of different biofilm communities. Adhesion to the surface provides considerable advantages such as protection against antimicrobial agents, acquisition of new genetic traits, and the nutrient availability and metabolic co-operability. Anthony van Leeuwenhoek, who discovered microbial attachment to his own tooth surface, is credited with the discovery of biofilm. The formation of biofilm takes place in three steps. Biofilm is responsible for chronic bacterial infection, infection on medical devices, deterioration of water quality and the contamination of food. This article provides an overview of the formation of biofilm, structure, role in microbial communities and its applications.

Keywords: Biofilm, polymeric substance, hydrodynamics, probes, pathogenesis

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Halotolerant microorganisms able to live in saline environments offer a multitude of actual or potential applications in various fields of biotechnology. This is why some strains of Halobacteria from an Algerian culture collection were screened for biosurfactant production in a standard medium using the qualitative drop-collapse test and emulsification activity assay. Five of the Halobacteria strains reduced the growth medium surface tension below 40 mN m^{-1} , and two of them exhibited high emulsion-stabilizing capacity. Diesel oil-in-water emulsions were stabilized over a broad range of conditions, from pH 2 to 11, with up to 35% sodium chloride or up to 25% ethanol in the aqueous phase. Emulsions were stable to three cycles of freezing and thawing. The components of the biosurfactant were determined; it contained sugar, protein and lipid. The two Halobacteria strains with enhanced biosurfactant producers, designated strain A21 and strain D21, were selected to identify by phenotypic, biochemical characteristics and by partial 16S rRNA gene sequencing. The strains have Mg^{2+} , and salt growth requirements are always above 15% (w/v) salts with an optimal concentration of 15–25%. Analyses of partial 16S rRNA gene sequences of the two strains suggested that they were halophiles belonging to genera of the family Halobacteriaceae, *Halovivax* (strain A21) and *Haloarcula* (strain D21). To our knowledge, this is the first report of biosurfactant production at such a high salt concentration.

Keywords: Halobacteria - Screening - Biosurfactant - Surface tension

Pollen Biotechnology

M Sahney and S. Chaurasia. (Department of Botany, University of Allahabad, Allahabad. Email: manjusahney@rediffmail.com). Pollen production in some allergenic plant TAXA of Allahabad. Indian Journal of Aerobiology, Volume 21 (2) (2008): 69 - 72.

The paper presents pollen production study of 15 selected plant taxa which are important pollen allergens of Allahabad. Among them highest pollen production per anther was estimated in *Cannabis saliva* followed by *Cassia fistula*. Other taxa in decreasing order of pollen production/anther were *Ailanthu excelsa*, *Holoptelea integrifolia*, *Putranjiva roxburghii*, *Argemone mexicana*, *Amaranthus spinosus*, *Amaranthus viridis*, *Chenopodium murale*, *Brassica compestris*, *Chenopodium album*, *Cynodon dactylon*, *Prosopis juliflora*, *Ricinus communis* and *Azadirachta indica*. Data of pollen production of investigated plant taxa have been correlated with the anther and pollen size and with their representation in the atmosphere of Allahabad during 2002 – 2004.

Key words: Pollen production, allergenic taxa, aerial incidence, mode of pollination.

Jon S. West¹, Simon D. Atkins¹, Jean Emberlin² and Bruce D.L. Fitt¹. (¹Rothamsted Research, Harpenden, Hertfordshire, AL5 2JQ, UK, ²National Pollen and Aerobiology Research Unit, University of Worcester, Henwick Grove, Worcester, WR2 6AJ, UK). PCR to predict risk of airborne disease. Trends in Microbiology, Volume 16(8) (2008): 380-387

Plant, animal and human diseases spread by microscopic airborne particles have had major economic and social impacts during history. Special air-sampling devices have been used to collect such particles since the 19th century but it has often been impossible to identify them accurately. Exciting new opportunities to combine air sampling with quantitative PCR to identify and count these particles are reviewed, using crop pathogen examples. These methods can be

used to predict the risk of unexpected outbreaks of airborne diseases by identifying increases in pathogen inoculum or genetic changes in pathogen populations that render control ineffective. The predictions can provide guidance to policymakers, health professionals or the agricultural industry for the development of strategies to minimise the risk of severe pandemics.

J. Douwes^a, W. Eduard^b and P.S. Thorne^c. (^aMassey University, Wellington, New Zealand, ^bNational Institute of Occupational Health, Oslo, Norway, ^cUniversity of Iowa, Iowa City, IA, USA). *Bioaerosols. International Encyclopedia of Public Health, Pages 287-297*

Bioaerosols are airborne compounds or microfragments from plant or animal matter or from microorganisms but also comprise whole microorganisms that are either dead or alive. Exposure to these agents may cause infectious diseases, allergic diseases, acute toxic effects, respiratory diseases, neurological effects, and possibly cancer. Respiratory symptoms and disease are the most common health effects associated with noninfectious bioaerosols and include asthma, hay fever, organic dust toxic syndrome, hypersensitivity pneumonitis, and chronic bronchitis. Bioaerosol exposure has also been demonstrated to adversely affect lung function and might play a role in sick building syndrome. Paradoxically, indoor exposure to moderate levels of bioaerosols has been suggested to reduce the risk of developing allergies and allergic asthma in early life. In this article we present an overview of the health effects associated with bioaerosol exposure in both the indoor and occupational environment. We also describe the major agents assumed to play a causal role in the development of bioaerosol-related health effects.

Author Keywords: Airflow obstruction; Allergens; Asthma; Bioaerosol; Chronic bronchitis; Endotoxin; Fungi; (1→3)-β-D-glucans; Hygiene hypothesis; Hypersensitivity pneumonitis; Infectious diseases; Mycotoxins; Organic dust toxic syndrome; Pulmonary hemorrhage; Rhinitis; Sick building syndrome

Linda D Stetzenbach, Mark P Buttner and Patricia Cruz. (Harry Reid Center for Environmental Studies, 4505 South Maryland Parkway, University of Nevada-Las Vegas, Las Vegas, Nevada 89154-4009, USA). *Detection and enumeration of airborne biocontaminants. Current Opinion in Biotechnology, Volume 15(3) (2004): 170-174*

The sampling and analysis of airborne microorganisms has received attention in recent years owing to concerns with mold contamination in indoor environments and the threat of bioterrorism. Traditionally, the detection and enumeration of airborne microorganisms has been conducted using light microscopy and/or culture-based methods; however, these analyses are time-consuming, laborious, subjective and lack sensitivity and specificity. The use of molecular methods, such as quantitative polymerase chain reaction amplification, can enhance monitoring strategies by increasing sensitivity and specificity, while decreasing the time required for analysis.

IAQ, indoor air quality; **IPC**, internal positive control; **PCR**, polymerase chain reaction; **QPCR**, quantitative polymerase chain reaction; **WMD**, weapons of mass destruction

Bruce Knox and Cenk Suphioglu. (Pollen and Allergen Research Group, School of Botany, The University of Melbourne, Parkville, Victoria 3052, Australia). *Environmental and molecular biology of pollen allergens. Trends in Plant Science, Volume 1(5) (1996): 156-164*

The release of pollen into the air is a normal part of the sexual cycle in many wind-pollinated plants. Unfortunately, however, certain pollen grains contain specific proteins or glycoproteins that can result in the familiar debilitating symptoms of hayfever and asthma in humans. This, together with the dramatic increase in the incidence of allergic disease in recent years, has led to increasing public concern about allergenic pollen. It is important to examine the distribution of pollen in the air, the particular molecular features of the allergens, and, perhaps most intriguingly, what role these highly interactive molecules play in pollen growth and development.

J. Lacey^{a, b} and J. Dutkiewicz^{a, b}. (^aInstitute of Arable Crops Research, Rothamsted Experimental Station, Harpenden, Herts AL5 2JQ, U.K., ^bInstitute of Agricultural Medicine, P.O. Box 185, ul. Jaczewskiego 2, 20-950 Lublin, Poland). Bioaerosols and occupational lung disease. *Journal of Aerosol Science*, Volume 25(8) (1994): 1371-1404

Bioaerosols are formed by suspension of particles of biological origin in the air. They come from a wide range of sources, many of which are associated with particular occupations. Many of the components have been implicated in occupational lung disease. The agents include viruses: bacteria; actinomycete, fungal, moss and fern spores; algal and plant cells; insects and mites and their fragments and excreta; proteins from plant and animal sources; enzymes, antibiotics and other products from biotechnological processes; endotoxins from Gram-negative bacteria; and mycotoxins and glucans from fungi. Infections from pathogenic viruses, bacteria and fungi may occur in some work environments but more often the symptoms encountered are of mucous membrane irritation, bronchitis and obstructive pulmonary disease, allergic rhinitis and asthma, allergic alveolitis (granulomatous pneumonitis) or organic dust toxic syndrome (inhalation fever or toxic pneumonitis). Exposure to bioaerosols may occur in many different occupations, especially those in which stored products are handled or where aerosols are created as a result of leaks from equipment intentionally or accidentally contaminated with microorganisms or during particular operations as, for instance, in laboratories and during post-mortem or surgical procedures. This article reviews the spectrum of agents involved in occupational lung disease, the work environments in which they occur, the characteristics of the diseases and their prevention.

W.D. Griffiths^a and G.A.L. DeCosemo^a. (^aWarren Spring Laboratory, Gunnels Wood Road, Stevenage, Herts SG1 2BX, U.K.). The assessment of bioaerosols: A critical review. *Journal of Aerosol Science*, Volume 25(8) (1994): 1425-1458

The main objective of this work is to review methods for the assessment of bioaerosols and consider some of the problems which arise. This paper investigates the main techniques currently available for sampling and detecting airborne microorganisms, examines a number of factors which can affect their survival, and discusses problems associated with the production of test bioaerosols. Ultimately it is hoped that a better understanding of these will provide a basis for the development of standard assessment protocols for bioaerosols and also help in the establishment of bioaerosol standards.

The need for aerobiological monitoring in occupational hygiene, containment of bioprocessing equipment, deliberate release of microorganisms, contamination of food and pharmaceutical products and general scientific research, has been identified. Despite the fact that aerobiological

monitoring has wide application and may be carried out to comply with guidelines or regulations there are, at present, no standard protocols for sampling airborne microorganisms. Six criteria, which need to be considered when carrying out any aerobiological monitoring, have been identified and defined.

Samplers used to collect bioaerosols are described and areas of their application highlighted. Few have been fully characterised in terms of their inlet or bioefficiencies. Samplers not currently used but which have potential use in bioaerosol monitoring have also been identified and described. A series of microbial assay methods are described in terms of their sensitivity, speed, and their relationship with the choice of sampler. The interdependence of sampling and assay methods is discussed.

Methods for the production of bioaerosols, from both liquid suspension and dry powder, are described. Techniques for the controlled production of poly and monodisperse aerosols are discussed. The survival characteristics of microorganisms are considered because they can greatly influence the effectiveness of sampling and assay methods. A number of recommendations and a programme of research work are outlined in order to further the understanding of the characteristics, behaviour, and assessment of bioaerosols prior to consideration of the possible establishment of bioaerosol standards for test purposes.

Biotechnology Policy Issue

Jennifer Gaudioso^a, Lisa Astuto Gribble^a and Reynolds M. Salerno^a. (^aInternational Biological Threat Reduction, Sandia National Laboratories Albuquerque, NM). *Biosecurity: Progress and Challenges. Journal of the Association for Laboratory Automation, Volume 14(3) (2009): 141-147*

Bioscience facilities are essential to the efforts to combat both naturally occurring infectious diseases and bioterrorism. But both the general public and policy makers are questioning how bioscience institutions address the safety and security risks of handling infectious disease causing organisms. As a result, new regulations at the national level in many countries and international initiatives from the United Nations, World Health Organization, and others are having direct consequences for the operation of bioscience. In particular, laboratory biosecurity is a relatively new and evolving paradigm for bioscience facilities, which have an obligation to ensure their facilities operate safely and securely. However, although progress has been made in these areas, numerous challenges remain throughout the world, and much work remains. It is the responsibility of both the scientific community and policy makers to work collaboratively to ensure responsible use of pathogens and toxins, equipment, and expertise.

Keywords: biosecurity; biosafety; biorisk; bioterrorism

Guillaume Gruère^a and Debdatta Sengupta^{1, a}. (^aEnvironment and Production Technology Division, International Food Policy Research Institute, 2033 K Street, Washington, DC 20006-1002, USA). *GM-free private standards and their effects on biosafety decision-making in developing countries. Food Policy, Available online May 2009 Article in Press,*

We provide a comprehensive review of international cases where GM-free private standards set up by food companies in developed countries have influenced biosafety policymaking in

developing countries. We find 29 cases where private importers have directly or indirectly affected policy decisions in 21 countries. Most of the cases relate irrational fear of export losses to excessively precautionary decisions. These cases are based on two generally misleading premises: the belief that Europe or Japan represents the only market for exports, and the perception that non-GM segregation is infeasible or prohibitively costly in all situations. Our study also demonstrates the importance of information asymmetries across countries and agents and the role of risk aversion in seemingly irrational decision-making. The combination of these four factors helps us explain why presumed but unproven expected commercial losses still represents a significant impediment to biosafety policymaking in developing countries.

Keywords: Genetically modified food; Private standards; Biosafety; International trade

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An essential step in the development of products based on biotechnology is an assessment of their potential economic impacts and safety, including an evaluation of the potential impact of transgenic crops and practices related to their cultivation on the environment and human or animal health. The purpose of this paper is to provide an assessment method to evaluate the impact of biotechnologies that uses quantifiable parameters and allows a comparative analysis between conventional technology and technologies using GMOs. This paper introduces a method to perform an impact analysis associated with the commercial release and use of genetically modified plants, the Assessment System GMP Method. The assessment is performed through indicators that are arranged according to their dimension criterion likewise: environmental, economic, social, capability and institutional approach. To perform an accurate evaluation of the GMP specific indicators related to genetic modification are grouped in common fields: genetic insert features, GM plant features, gene flow, food/feed field, introduction of the GMP, unexpected occurrences and specific indicators. The novelty is the possibility to include specific parameters to the biotechnology under assessment. In this case by case analysis the factors of moderation and the indexes are parameterized to perform an available assessment.

Keywords: Biotechnology impacts; Genetically modified plants rapid appraisal; Impact assessment; Technological forecasting

Ian K. Dawson^a, Peter E. Hedley^b, Luigi Guarino^c and Hannah Jaenicke^a. (^aThe International Centre for Underutilised Crops, P.O. Box 2075, Colombo, Sri Lanka, ^bProgramme of Genetics, SCRI, Invergowrie, Dundee DD2 5DA, UK, ^cThe Global Crop Diversity Trust, c/o FAO, Viale delle Terme di Caracalla, 00153 Rome, Italy). **Does biotechnology have a role in the promotion of underutilised crops? [✉] Food Policy, Article in Press**

Rapidly developing biotechnology applications aimed at improving major crops receive large investments and could, in theory, play a role in the promotion of underutilised plant species in the tropics and subtropics, in order to address current and emerging challenges for agriculture. The application of such methods is, however, sometimes controversial, and the frequently considerable costs involved must be weighed against the limited resources available to develop underutilised species, as well as against the many alternative methods available for promotion. Through database searches, we take an evidence-based approach to assess whether there are clear examples where biotechnology has been used practically to enhance the cultivation of underutilised plants at a field level. We conclude that tissue culture and micropropagation techniques have proven useful, but for other applications benefits are generally unclear at present, although ongoing work suggests genomic and genetic modification approaches may in future be significant for a subset of underutilised species. Successful outcomes, however, appear to be limited by a lack of integrated thinking during the use of biotechnology methods. We review the particular limitations and risks associated with applying biotechnology to underutilised crops, including the negative consequences of technology centralisation. In addition, the specific actions needed to ensure that smallholder farmers in low-income countries better benefit during the use of biotechnology on underutilised species, by placing a stronger emphasis on partnerships and by proper monitoring of benefits along value chains, are described.

Keywords: Biotechnology; Integration; Practical application; Underutilised plants

Agricultural Biotechnology

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In the present study the natural abundance of ¹³C is quantified in agricultural soils in Mexico which have been submitted to different agronomic practices, zero and conventional tillage, retention of crop residues (with and without) and rotation of crops (wheat and maize) for 17 years, which have influenced the physical, chemical and biological characteristics of the soil. The natural abundance of C13 is quantified by near infrared spectra (NIRS) with a remote reflectance fibre optic probe, applying the probe directly to the soil samples.

Discriminate partial least squares analysis of the near infrared spectra allowed to classify soils with and without residues, regardless of the type of tillage or rotation systems used with a prediction rate of 90% in the internal validation and 94% in the external validation. The NIRS calibration model using a modified partial least squares regression allowed to determine the $\delta^{13}\text{C}$ in soils with or without residues, with multiple correlation coefficients 0.81 and standard error

prediction 0.5‰ in soils with residues and 0.92 and 0.2‰ in soils without residues. The ratio performance deviation for the quantification of $\delta^{13}\text{C}$ in soil was 2.5 in soil with residues and 3.8 without residues. This indicated that the model was adequate to determine the $\delta^{13}\text{C}$ of unknown soils in the -16.2‰ to -20.4‰ range. The development of the NIR calibration permits analytic determinations of the values of $\delta^{13}\text{C}$ in unknown agricultural soils in less time, employing a non-destructive method, by the application of the fibre optic probe of remote reflectance to the soil sample.

Keywords: $\delta^{13}\text{C}$; Soil; Near infrared reflectance spectroscopy (NIRS)

Mikhail A. Semenov^a, Pierre Martre^{b, c} and Peter D. Jamieson^d. (^aCentre for Mathematical and Computational Biology, Rothamsted Research, Harpenden, Herts, AL5 2JQ, UK, ^bINRA, UMR1095 Genetic, Diversity and Ecophysiology of Cereals, Clermont-Ferrand, F-63 100, France, ^cUniversité Blaise Pascal, UMR1095 Genetic, Diversity and Ecophysiology of Cereals, Clermont-Ferrand, F-63 100, France, ^dNew Zealand Institute for Crop & Food Research, Private Bag 4704, Christchurch, New Zealand). **Quantifying effects of simple wheat traits on yield in water-limited environments using a modelling approach. *Agricultural and Forest Meteorology*, Volume 149(6-7) (2009): 1095-1104**

Availability of water for plant growth is a key factor determining plant distribution in natural ecosystems and is the most important limiting factor in agricultural systems. The high environmental and economical cost of irrigation, required to maintain grain yields in water scarce environments, gives an incentive for improvements in water use efficiency of the crop. The objective of our study is to quantify the effects of changes in simple component plant traits on wheat yield under limited water supplies using a modelling approach. The Sirius wheat simulation model was used to perform analyses at two contrasting European sites, Rothamsted, UK and Seville, Spain, which represent major wheat growing areas in these countries. Several physiological traits were analysed to explore their effects on yield, including drought avoidance traits such as those controlling wheat development (phyllochron and grain filling duration), canopy expansion (maximum surface area of culm leaves) and water uptake (root vertical expansion rate and efficiency of water extraction) and drought tolerance traits such as responses of biomass accumulation and leaf senescence to water stress. Changes in parameters that control the effect of water stress on leaf senescence and biomass accumulation had the largest impact on grain yield under drought. The modified cultivar produced up to 70% more yield compared with the control for very dry years. Changes in phenology parameters, phyllochron and grain filling duration, did not improve yields at either site, suggesting that these parameters have been already optimised for climates in the UK and Spain through the breeding process. Our analysis illustrates the power of modelling in exploring and understanding complex traits in wheat. This may facilitate genetic research by focusing on experimental studies of component traits with the highest potential to influence crop performance.

Keywords: Crop improvement; Deconvoluting complex traits; $G \times E$ interactions; Crop simulation model; Sirius

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Gembloux, Belgium, ^bDepartment of Biotechnology, Walloon Agricultural Research Centre, Chaussé de Charleroi, 234, B-5030 Gembloux, Belgium). Role of *myo*-inositol phosphate synthase and sucrose synthase genes in plant seed development. *Gene*, Volume 439(1-2) (2009): 1-10

The aim of this review is to highlight the role of *myo*-inositol phosphate synthase (MIPS), which catalyses the first step in inositol biosynthesis and of sucrose synthase (Sus), an enzyme involved in UDP-glucose formation, the principal nucleoside diphosphate in the sucrose cleavage reaction and in trehalose biosynthesis. These two enzymes are involved in various physiological processes including seed growth and resistance to biotic and abiotic stresses.

The study of mutated *MIPS* and *Sus* genes in some crops, such as soybean and cotton, has shown that these two proteins are directly involved in embryogenesis. They exhibit several isoforms that are essential for normal seed development.

The possible role of both genes in seed development is discussed in this review.

Keywords: *myo*-inositol phosphate synthase (MIPS); Sucrose synthase (Sus); Seed development

Abbreviations: EMS, ethyl methanesulfonate; EST, express sequences tag; IAA, indole-3-acetic acid; LPA, low phytic acid; MIPS, *myo*-inositol phosphate synthase; P-Sus, particulate Sus; SSH, suppressive subtractive hybridization; S-Sus, soluble Sus; Sus, sucrose synthase; T6P, trehalose-6-phosphate; TPP, trehalose-6-phosphate phosphatase; TPS, trehalose-6-phosphate synthase; UDP-glucose, uridine-5-diphosphoglucose

Eun-Hee Doo^a, Won-Heong Lee^b, Hyo-Seel Seo^a, Jin-Ho Seo^b and Jin-Byung Park^a. (^aDepartment of Food Science & Engineering, Ewha Womans University, Seoul 120-750, Republic of Korea, ^bDepartment of Agricultural Biotechnology and Center for Agricultural Biomaterials, Seoul National University, Seoul 151-921, Republic of Korea). Productivity of cyclohexanone oxidation of the recombinant *Corynebacterium glutamicum* expressing *chnB* of *Acinetobacter calcoaceticus*. *Journal of Biotechnology*, Volume 142(2) (2009): 164-169

The biocatalytic efficiency of recombinant *Corynebacterium glutamicum* expressing the *chnB* gene encoding cyclohexanone monooxygenase (CHMO) of *Acinetobacter calcoaceticus* NCIMB 9871 was investigated. Optimization of an expression system and induction conditions enabled the recombinant biocatalyst to produce CHMO to a specific activity of ca. 0.5 U mg⁻¹ protein. Tight control of feeding of an energy source (i.e., glucose) and dissolved oxygen tension during fed-batch culture-based biotransformation allowed the cells to produce ϵ -caprolactone to a concentration of 16.0 g l⁻¹. The specific and volumetric productivity for cyclohexanone oxidation were 0.12 g g dry cells⁻¹ h⁻¹ (17.5 U g⁻¹ of dry cells) and 2.3 g l⁻¹ h⁻¹ (330 U l⁻¹), respectively. These values correspond to over 5.4- and 2.7-fold of recombinant *Escherichia coli* expressing the same gene under similar reaction conditions. It could be concluded that the recombinant *C. glutamicum* is a promising biocatalyst for Baeyer–Villiger oxidations.

Keywords: Biocatalysis; Baeyer–Villiger oxidation; *Corynebacterium glutamicum*; Cyclohexanone monooxygenase; ϵ -Caprolactone

Bioenergy

Novy Srihartati Kasim^a, Tsung-Han Tsai^a, Setiyo Gunawan^{a, b} and Yi-Hsu Ju^a. (Department of Chemical Engineering, National Taiwan University of Science and Technology, 43 Sec.4, Keelung Road, Taipei 106-07, Taiwan, ^bDepartment of Chemical Engineering, Institut Teknologi Sepuluh Nopember, Kampus ITS Keputih Sukolilo, Surabaya 60111, Indonesia). Biodiesel production from rice bran oil and supercritical methanol. *Bioresource Technology*, Volume 100(8) (2009): 2399-2403

In this study, production of biodiesel from low cost raw materials, such as rice bran and dewaxed-degummed rice bran oil (DDRBO), under supercritical condition was carried out. Carbon dioxide (CO₂) was employed as co-solvent to decrease the supercritical temperature and pressure of methanol. The effects of different raw materials on the yield of biodiesel production were investigated. In situ transesterification of rice bran with supercritical methanol at 30 MPa and 300 °C for 5 min was not a promising way to produce biodiesel because the purity and yield of fatty acid methyl esters (FAMES) obtained were 52.52% and 51.28%, respectively. When DDRBO was reacted, the purity and yield were 89.25% and 94.84%, respectively. *Trans*-FAMES, which constituted about 16% of biodiesel, were found. They were identified as methyl elaidate [*trans*-9], methyl linoleaidate [*trans*-9, *trans*-12], methyl linoleaidate [*cis*-9, *trans*-12], and methyl linoleaidate [*trans*-9, *cis*-12]. Hydrocarbons, which constituted about 3% of the reaction product, were also detected.

Keywords: In situ transesterification; Rice bran; Rice bran oil; Supercritical methanol

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This paper presents the results of experiments to test biodiesel waste (glycerine – g-phase) as an organic carbon source for the removal of nitrate in a WWTP denitrification process. Investigation of g-phase was first centered on g-phase utilization as an external source for denitrification under laboratory conditions and consequently, after positive results from the laboratory investigation, g-phase was applied in the denitrification process in the WWTP Vrútky (35,000 PE). This WWTP had insufficient nitrogen removal via denitrification. Denitrification was insufficient due to an influent with a low BOD₅/N ratio (1.7:1) entering into the activated sludge tank. Laboratory experiments and calculations showed that, to reach N_{total} concentration under 10 mg l⁻¹ in effluent, a biodiesel waste dose of 500 kg_{COD} d⁻¹ was necessary. Glycerol phase (g-phase) dosing into the denitrification tank increased denitrification efficiency by 2.0 – 5.0 mg_{NO₃-N} l⁻¹ per 100 l of g-phase dose into the denitrification tank.

Keywords: Biodiesel; Denitrification process; External organic carbon; Glycerol; Nitrogen removal

Nadir Dizge^a, Coskun Aydiner^a, Derya Y. Imer^a, Mahmut Bayramoglu^b, Aziz Tanriseven^c and Bülent Keskinler^a. (^aGebze Institute of Technology, Department of Environmental Engineering, Gebze 41400, Turkey, ^bGebze Institute of Technology, Department of Chemical Engineering, Gebze 41400, Turkey, ^cGebze Institute of Technology, Department of Biochemistry, Gebze 41400, Turkey). **Biodiesel production from sunflower, soybean, and waste cooking oils by transesterification using lipase immobilized onto a novel microporous polymer. *Bioresource Technology*, Volume 100(6) (2009): 1983-1991**

This study aims at carrying out lipase-catalyzed synthesis of fatty acid methyl esters (biodiesel) from various vegetable oils using lipase immobilized onto a novel microporous polymeric matrix (MPPM) as a low-cost biocatalyst. The research is focused on three aspects of the process: (a) MPPM synthesis (monolithic, bead, and powder forms), (b) microporous polymeric biocatalyst (MPPB) preparation by immobilization of lipase onto MPPM, and (c) biodiesel production by MPPB. Experimental planning of each step of the study was separately carried out in accordance with design of experiment (DoE) based on Taguchi methodology.

Microporous polymeric matrix (MPPM) containing aldehyde functional group was synthesized by polyHIPE technique using styrene, divinylbenzene, and polyglutaraldehyde. *Thermomyces lanuginosus* lipase was covalently attached onto MPPM with 80%, 85%, and 89% immobilization efficiencies using bead, powder, and monolithic forms, respectively. Immobilized enzymes were successfully used for the production of biodiesel using sunflower, soybean, and waste cooking oils. It was shown that immobilized enzymes retain their activities during 10 repeated batch reactions at 25 °C, each lasting 24 h. Since the developed novel method is simple yet effective, it could have a potential to be used industrially for the production of chemicals requiring immobilized lipases.

Keywords: Biodiesel; Transesterification; Microporous polymeric biocatalyst; *Thermomyces lanuginosus* lipase; Taguchi methodology

Abderrahim Bouaid^a, Mercedes Martinez^a and Jose Aracil^a. (^aChemical Engineering Department, Faculty of Chemistry, University of Complutense, 28040 Madrid, Spain). **Production of biodiesel from bioethanol and *Brassica carinata* oil: Oxidation stability study. *Bioresource Technology*, Volume 100(7) (2009): 2234-2239**

In the present work the synthesis from bioethanol and *Brassica carinata*, as alternative vegetable oil, using KOH as catalyst, has been developed and optimized by application of the factorial design and response surface methodology (RSM). Temperature and catalyst concentration were found to have significant influence on conversion. A second-order model was obtained to predict conversions as a function of temperature and catalyst concentration. The maximum yield of ester (98.04%) was obtained working with an initial concentration of catalyst (1.5%) and an operation temperature of (35 °C). Results show that the acid value, peroxide value, and viscosity, increased while the iodine value decreased with increasing storage time of the biodiesel sample. Fatty acid ethyl esters (biodiesel) from *B. carinata* oil were very stable because they did not demonstrate rapid increase in peroxide value, acid value, and viscosity with increasing storage time to a period of 12 months.

Keywords: Biodiesel; *Brassica carinata* oil; Fatty acid ethyl esters; Response surface methodology (RSM); Oxidation stability

T. Schleicher^a, R. Werkmeister^a, W. Russ^a and R. Meyer-Pittroff^b. (^aInstitute of Resource and Energy Technology, Technische Universität München, Weihenstephaner Steig 22, 85350 Freising, Germany, ^bCompetence Pool Weihenstephan, Technische Universität München, Weihenstephaner Steig 23, 85350 Freising, Germany). **Microbiological stability of biodiesel–diesel-mixtures. Bioresource Technology, Volume 100(2) (2009): 724-730**

The biodegradation of rapeseed oil methyl ester (RME) in pure and in mixtures with diesel fuel was investigated. Higher ratio of diesel fuel in the mixture resulted in higher count of bacteria. Fungal growth was advanced by higher RME contents. The growth of microorganisms gained from soil was strongest in B 20 (20 vol.% biodiesel and 80 vol.% diesel fuel) mixtures followed by B 5 (5 vol.% biodiesel and 95 vol.% diesel fuel) mixtures and pure RME. The formation of free fatty acids (FFA) in the RME sample was measured according to DIN EN 14214. The content of FFA in inoculated RME samples rose from 0.08 mass% to 0.344 mass% at the beginning. The oxidation stability of inoculated samples of B 20, B 5 and pure RME decreased faster than the oxidation stability of blank samples. An optical evaluation showed the formation of turbidity. Partly, the formation of sediment was observed, especially in B 20 and B 5 samples.

Keywords: Diesel; Biodiesel; Biodegradation; Microbial growth

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The objective of this work was to study the synthesis of ethyl esters via esterification of soybean oil deodorizer distillate with ethanol, using solid acid catalysts and commercial immobilized lipases, in a solvent-free system. Three commercially immobilized lipases were used, namely, Lipozyme RM-IM, Lipozyme TL-IM, and Novozym 435, all from Novozymes. We aimed for optimum reaction parameters: temperature, enzyme concentration, initial amount of ethanol, and its feeding technique to the reactor (stepwise ethanolysis). Reaction was faster with Novozym 435. The highest conversion (83.5%) was obtained after 90 min using 3 wt.% of Novozym 435 and two-stage stepwise addition of ethanol at 50Å°C. Four catalysts were also tested: zeolite CBV-780, SAPO-34, niobia, and niobic acid. The highest conversion (30%) was obtained at 100Å°C, with 3 wt.% of CBV-780 after 2.5 h. The effects of zeolite CBV 780 concentration were studied, resulting in a conversion of 49% using 9 wt.% of catalyst.

Keywords: Esterification - SODD - Biodiesel - Ethanol - Immobilized lipase - Zeolite

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dong, Ansung-city, Kyonggi-do, 456-749, Korea). Biodiesel Production from Various Oils Under Supercritical Fluid Conditions by Candida antarctica Lipase B Using a Stepwise Reaction Method. Applied Biochemistry and Biotechnology, Volume 156(1-3) (2009): 24-34

In this study, we evaluate the effects of various reaction factors, including pressure, temperature, agitation speed, enzyme concentration, and water content to increase biodiesel production. In addition, biodiesel was produced from various oils to establish the optimal enzymatic process of biodiesel production. Optimal conditions were determined to be as follows: pressure 130 bar, temperature 45 °C, agitation speed 200 rpm, enzyme concentration 20%, and water contents 10%. Among the various oils used for production, olive oil showed the highest yield (65.18%) upon transesterification. However, when biodiesel was produced using a batch system, biodiesel conversion yield was not increased over 65%; therefore, a stepwise reaction was conducted to increase biodiesel production. When a reaction medium with an initial concentration of methanol of 60 mmol was used and adjusted to maintain this concentration of methanol every 1.5 h during biodiesel production, the conversion yield of biodiesel was 98.92% at 6 h. Finally, reusability was evaluated using immobilized lipase to determine if this method was applicable for industrial biodiesel production. When biodiesel was produced repeatedly, the conversion rate was maintained at over 85% after eight reuses.

Keywords: Biodiesel - Initial reaction rate - Lipase activity - Optimization - Solubility - Supercritical fluid condition

Ayhan Demirbas^a. (^aSila Science, Trabzon, Turkey). Production of biodiesel fuels from linseed oil using methanol and ethanol in non-catalytic SCF conditions. Biomass and Bioenergy, Volume 33(1) (2009): 113-118

Methyl and ethyl esters as biodiesel fuels were prepared from linseed oil with transesterification reaction in non-catalytic supercritical fluids conditions. Biodiesel fuel is a renewable substitute fuel for petroleum diesel fuel made from vegetable or animal fats. Biodiesel fuel has better properties than that of petroleum diesel fuel such as renewable, biodegradable, non-toxic, and essentially free of sulfur and aromatics. The purpose of the transesterification process is to lower the viscosity of the oil. The viscosity values of linseed oil methyl and ethyl esters highly decreases after transesterification process. The viscosity values of vegetable oils vary between 27.2 and 53.6 mm² s⁻¹, whereas those of vegetable oil methyl esters between 3.59 and 4.63 mm² s⁻¹. Compared with no. 2 diesel fuel, all of the vegetable oil methyl esters were slightly viscous. The flash point values of vegetable oil methyl esters are highly lower than those of vegetable oils. The transesterification of linseed oil in supercritical fluids such as methanol and ethanol has proved to be the most promising process. Methanol is the commonly used alcohol in this process, due in part to its low cost. Methyl esters of vegetable oils have several outstanding advantages among other new-renewable and clean engine fuel alternatives. The most important variables affecting the methyl ester yield during the transesterification reaction are molar ratio of alcohol to vegetable oil and reaction temperature. Biodiesel has become more attractive recently because of its environmental benefits. Biodiesel is an environmentally friendly fuel that can be used in any diesel engine without modification.

Keywords: Linseed oil; Biodiesel; Viscosity; Transesterification; Methanol; Ethanol

Name of Journals

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2. Aerobiologia
3. Annual Review-Plant Pathology
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