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Environmental Information System
INDIA

ENVIS CENTER
on
ENVIRONMENTAL BIOTECHNOLOGY

Abstract Vol. XVI

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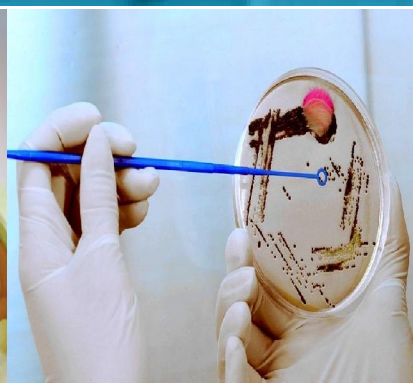

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ENVIS CENTRE

on

ENVIRONMENTAL BIOTECHBNOLOGY

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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 biannualy Abstract Volume on our thematic area Environmental Biotechnology under fourteen subheads. The volume contains abstracts of scientific articles from relavent 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 16th 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, 2010. 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 varity 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 warm. 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 varity of environments. These specific substances may include sugars, proteins, or humas and varity 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 allerginicity 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 ecofreindly 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	Corpn	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	Ecotoxico	Ecotoxicology
Biotech	Biotechnique(s)	Endocrinol	Endocrinological
Biotechno	Biotechnology	Engg	Engineering
Biotechnol	Bitechnological	Engrs	Engineers
Bldg	Building	Env	Environment
Bot	Botany	Environ	Environmental
Botl	Botanical	Epidemic	Epidemiology
Br	Branch	Epidemiol	Epidemiological
Bull	Bulletin	Estd	Establishment
Cent	Centre	Ethnopharmaco	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
Geogrl	Geographical	Occ	Occassional
Geol	Geological	Occupl	Occupational
Geosci	Geoscience	Oceanogr	Oceanogoraphy
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	Immunlogical	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

Sandro Froehner[1] and Marcell Maceno[1]. ([1]Departamento De Engenharia Ambiental, Centro PolitÃ©cnico, Universidade Federal Do ParanÃ¡, Curitiba, ParanÃ¡, 81531-990, Brazil). Assessment of bioaccumulation of biphenyls in the trophic chain of a coastal area of Parana, Brazil. Environmental Monitoring and Assessment, Volume 164(1-4) (2010): 189-198

The presence of biphenyl was investigated in sediments and water in Paranagua Bay. Chemicals compounds like biphenyl can cause several effects on the ecosystems such as bioaccumulation. Biphenyl and similar compounds are subject to bioaccumulation, which in turn may harm the local ecosystem. The bioaccumulation in the local trophic chain was evaluated using a mathematical model based on toxicokinetic properties of the compound in the organisms. The results showed that even in water, the concentration of biphenyl was high, 0.82 ng/L. Also, in the fishes, the concentrations calculated by the model were higher than the maximum than the maximum allowed for human consumption.

Keywords: Bioaccumulation - Ecological effects - Persistent organic pollutants

Charles Hofer[a], Frank J. Gallagher[a] and Claus Holzapfel[b]. ([a]Department of Ecology, Evolution and Natural Resources, Rutgers, the State University of New Jersey, 14 College Farm Rd., New Brunswick, NJ 08901-8551, USA, [b] Department of Biological Sciences, Rutgers, the State University of New Jersey, Newark, 195 University Ave., Newark, NJ 07102-1811, USA). Metal accumulation and performance of nestlings of passerine bird species at an urban brownfield site. Environmental Pollution, Volume 158(5) (2010): 1207-1213

The use of passerine species as bioindicators of metal bioaccumulation is often underutilized when examining the wildlife habitat value of polluted sites. In this study we tested feathers of nestlings of two common bird species (house wren and American robin) for accumulation of Pb, Zn, As, Cr, Cu, Fe in comparison of a polluted, urban brownfield with a rural, unpolluted site. House wren nestlings at the study site accumulated significantly greater concentrations of all target metals except Zn. At the polluted site we found significant species differences of metal concentrations in feathers, with house wrens accumulating greater concentrations of Pb, Fe, and Zn but slightly lesser accumulations of Cr and Cu than American robins. Although house wren nestlings demonstrated significant accumulation of metals, these concentrations showed little effect on size metrics or fledge rates during the breeding season compared to nestlings from the control site.

Nestlings of birds in an urban brownfield accumulated soil contaminants but did not show signs of reduced breeding success or growth.

Keywords: Bioindicators; Metals; Restoration ecology; Urban ecology

I. Del Toro[a], K. Floyd[b], J. Gardea-Torresdey[c] and D. Borrok[d]. (a Department of Organismic and Evolutionary Biology, University of Massachusetts at Amherst, 611 N.

Pleasant Street Amherst, MA 01003, USA, b Environmental Science and Engineering, University of Texas at El Paso, 500 W. University Ave., El Paso, TX 79968, USA, c Department of Chemistry, University of Texas at El Paso, 500 W. University Ave., El Paso, TX 79968, USA, d Department of Geological Sciences, University of Texas at El Paso, 500 W. University Ave., El Paso, TX 79968, USA). Heavy metal distribution and bioaccumulation in Chihuahuan Desert Rough Harvester ant (Pogonomyrmex rugosus) populations. Environmental Pollutionm Volume 158(5) (2010): 1281-1287

Heavy metal contamination can negatively impact arid ecosystems; however a thorough examination of bioaccumulation patterns has not been completed. We analyzed the distribution of As, Cd, Cu, Pb and Zn in soils, seeds and ant (*Pogonomyrmex rugosus*) populations of the Chihuahuan Desert near El Paso, TX, USA. Concentrations of As, Cd, Cu, and Pb in soils, seeds and ants declined as a function of distance from a now inactive Cu and Pb smelter and all five metals bioaccumulated in the granivorous ants. The average bioaccumulation factors for the metals from seeds to ants ranged from 1.04× (As) to 8.12× (Cd). The findings show bioaccumulation trends in linked trophic levels in an arid ecosystem and further investigation should focus on the impacts of heavy metal contamination at the community level. Heavy metals bioaccumulate in desert ants.

Keywords: Granivory; Hymenoptera Formicidae; Bioaccumulation; Heavy metals

Ying Lua[b], Fei Donga, Claire Deaconb, Huo-jun Chena, Andrea Raabb[c] and Andrew A. Mehargb. ([a] College of Natural Resources and Environment, South China Agricultural University, Guangzhou 510642, China, [b] School of Biological Science, University of Aberdeen, Aberdeen AB24 3UU, UK, [c] Department of Chemistry, University of Aberdeen, Aberdeen AB24 3UE, UK). Arsenic accumulation and phosphorus status in two rice (Oryza sativa L.) cultivars surveyed from fields in South China. Environmental Pollution, Volume 158(5) (2010): 1536-1541

The consumption of paddy rice (*Oryza sativa* L.) is a major inorganic arsenic exposure pathway in S.E. Asia. A multi-location survey was undertaken in Guangdong Province, South China to assess arsenic accumulation and speciation in 2 rice cultivars, one an *Indica* and the other a *hybrid Indica*. The results showed that arsenic concentrations in rice tissue increased in the order grain < husk < straw < root. Rice grain arsenic content of 2 rice cultivars was significant different and correlated with phosphorus concentration and molar ratio of P/As in shoot, being higher for the *Indica* cultivar than for the *hybrid Indica*, which suggests altering shoot phosphorus status as a promising route for breeding rice cultivars with reduced grain arsenic. Speciation of grain arsenic, performed using HPLC-ICP-MS, identified inorganic arsenic as the dominant arsenic species present in the rice grain.

Altering rice shoot phosphorus status is a promising route for breeding rice cultivars with reduced grain arsenic.

Keywords: Arsenic; China; Grain; Phosphorus; Rice

B. Duarte[a], M. Caetanoc, P.R. Almeida[a], b, C. Valec and I. Caçador[a]. (a Centro de Oceanografia, Instituto de Oceanografia, Campo Grande, 1749-1016 Lisboa, Portugal, b Departamento de Biologia, Universidade de Évora, Largo dos Colegiais 2, 7004-516 Évora, Portugal, c INRB/IPIMAR – Instituto Nacional de Recursos Biológicos, Av. Brasília, 1449-

006 Lisboa, Portugal). Accumulation and biological cycling of heavy metal in four salt marsh species, from Tagus estuary (Portugal). Environmental Pollution, Volume 158(5) (2010): Pages 1661-1668

Pools of Zn, Cu, Cd and Co in leaf, stem and root tissues of *Sarcocornia fruticosa*, *Sarcocornia perennis*, *Halimione portulacoides* and *Spartina maritima* were analyzed on a bimonthly basis, in a Tagus estuary salt marsh. All the major concentrations were found in the root tissues, being the concentrations in the aboveground organs neglectable for sediment budget proposes, as seen by the low root-aboveground translocation. Metal annual accumulation, root turnovers and cycling coefficients were also assessed. *S. maritima* showed the higher root turnovers and cycling coefficients for most of the analyzed metals, making this a phytostabilizer specie. By contrast the low root turnover, cycling coefficient and low root necromass generation makes *S. perennis* the most suitable specie for phytoremediation processes. Although the high amounts of metal return to the sediments, due to root senescence, salt marshes can still be considered sinks of heavy metals, cycling heavy metals mostly between sediment and root.

The efficiency of the phytoremediative processes and metal budgets are greatly influenced by the turnover periods and necromass generation.

Keywords: Metal cycling; Salt marshes; Root; Turnover periods

Jessica M. Reichmuth[a], Peddrick Weis[b], and Judith S. Weis[a]. ([a] Department of Biological Sciences, Rutgers, The State University of New Jersey, 195 University Avenue, Newark, NJ 07102, USA, [b] Department of Radiology, UMDNJ-The Medical School of New Jersey, Newark, NJ 07101, USA). Bioaccumulation and depuration of metals in blue crabs (Callinectes sapidus Rathbun) from a contaminated and clean estuary. Environmental Pollution, Volume 158(2) (2010): 361-368

Blue crabs from a contaminated estuary (Hackensack Meadowlands-HM) and a cleaner reference site (Tuckerton-TK) were analyzed for Cr, Cu, Hg, Pb, and Zn in muscle and hepatopancreas. Crabs from each site were taken into the laboratory and fed food from the other site, or in another experiment, transplanted to the other site for eight weeks. All crabs were analyzed for metals. Overall, tissue concentrations reflected environmental conditions. Tissue differences were found for Cu, Pb and Zn (all higher in hepatopancreas), and Hg (higher in muscle). HM muscle had more Hg than TK muscle, but did not decrease after transplanting or consuming clean food. HM crabs lost Cu, Pb and Zn in hepatopancreas after being fed clean food or transplanted. TK crabs increased Hg in muscle and Cr and Zn in hepatopancreas after transplantation or being fed contaminated (HM) food. Concentrations were variable, suggesting that blue crabs may not be fully reliable bioindicators of polluted systems.

The accumulation of metals within the muscle and hepatopancreas of blue crabs was highly variable, but often followed environmental concentrations.

Keywords: Bioaccumulation; *Callinectes sapidus*; Depuration; Metals; Pollution

Ofelia Dolores Hernández [1], Ángel José Gutiérrez [1], Dailos González-Weller [2] *, Gonzalo Lozano [3], Enrique García Melón [4], Carmen Rubio 1, Arturo Hardisson [1]. ([1]Department of

Public Health and Toxicology, University of La Laguna, 38071, La Laguna, Tenerife, Canary Islands, Spain, 2Health Inspection and Laboratory Service, Canary Health Service, 38006, Santa Cruz de Tenerife, Tenerife, Canary Islands, Spain, [3]Department of Animal Biology (Marine Sciences), University of La Laguna, 38206, La Laguna, Tenerife, Canary Islands, Spain, [4]Department of Navigation Sciences and Techniques, Technical College of Naval Seamanship, Machinery, Radios and Electronics, University of La Laguna, 38001, Santa Cruz de Tenerife, Tenerife, Canary Islands, Spain). Accumulation of toxic metals (Pb and Cd) in the sea urchin Diadema aff. antillarum Philippi, 1845, in an oceanic island (Tenerife, Canary Islands). Environmental Toxicology, Volume 25(3) (2010): 227 – 233

This document shows the results obtained from a study on the concentration of toxic heavy metals in the internal tissue and exoskeleton of sea urchins, collected from their natural habitat. The levels of lead and cadmium were measured by Graphite Furnace Atomic Absorption Spectrometry. The mean concentrations of lead and cadmium in the internal tissue were 304.04 and 260.54 µg/kg respectively, whereas in the shell they were 185.02 and 142.48 µg/kg. We also performed a statistical analysis of the differences in the distribution of metals between their exoskeleton and their internal content, a correlation study of the metal content in internal tissue and shell and sampling areas, and a correlation study between the metal content and sample size. Since the sea urchin *Diadema antillarum* presents a wide range of variation in metal content, this study suggests that this species is an excellent bioindicator of heavy metal contamination

Keywords: sea urchins • Canary Islands • lead and cadmium • graphite furnace atomic absorption spectrophotometry • accumulation • environmental bioindicator

Yin-Ming Kuo[1, 2], Maria S. SepÃºlveda[1, 3], Inez Hua[1, 4], Hugo G. Ochoa-AcuÃ±a[1, 5] and Trent M. Sutton[6]. ([1]School of Civil Engineering, Purdue University, 550 Stadium Mall Dr., West Lafayette, IN 47907, USA, [2]*Present address:* Department of Civil and Environmental Engineering, University of Vermont, 33 Colchester Ave., Burlington, VT 05405, USA, [3]Department of Forestry and Natural Resources, Purdue University, 195 Marsteller St., West Lafayette, IN 47907, USA, [4]Division of Environmental and Ecological Engineering, Purdue University, 500 Central Dr., West Lafayette, IN 47907, USA, [5]Department of Comparative Pathobiology, Purdue University, 725 Harrison St., West Lafayette, IN 47907, USA, [6]School of Fisheries and Ocean Sciences, University of Alaska Fairbanks, 245 O'Neill Building, Fairbanks, AK 99775, USA). Bioaccumulation and biomagnification of polybrominated diphenyl ethers in a food web of Lake Michigan. Ecotoxicology, Volume 19(4) (2010): 623-634

Polybrominated diphenyl ethers are hydrophobic chemicals and can biomagnify in food chains. Little is known about the biomagnification of PBDEs in the Lake Michigan food web. Plankton, *Diporeia*, lake whitefish, lake trout, and Chinook salmon were collected from Lake Michigan in 2006 between April and August. Fish liver and muscle and whole invertebrates were analyzed for six PBDEs (BDE-47, 99, 100, 153, 154, and 209). Carbon and nitrogen stable isotope ratios ($\delta^{13}C$ and $\delta^{15}N$) were also quantified in order to establish the trophic structure of the food web. Geometric means of ∑PBDE concentrations in fish ranged from 0.562 to 1.61 µg/g-lipid. BDE-209 concentrations ranged from 0.184 to 1.23 µg/g-lipid in all three fish species. ∑BDE-47, 99, and 209 comprised 80–94% of ∑PHDE molar concentration. Within each fish species, there were no significant differences in PBDE concentrations between liver and muscle. The highest concentration of BDE-209 (144 µg/g-lipid) was detected in *Diporeia*. Based on analysis of $\delta^{15}N$

and PBDE concentrations, BDE-47 and 100 were found to biomagnify, whereas BDE-209 did not. A significant negative correlation between BDE-209 and trophic level was found in this food web. Biomagnification factors were also calculated and again BDE-47 and 100 biomagnified between food web members whereas BDE-209 did not. *Diporeia* could be one of the main dietary sources of BDE-209 for fish in Lake Michigan; BDE-47 and 100 biomagnified within this food chain; the concentration of BDE-209 decreased at higher trophic levels, suggesting partial uptake and/or biotransformation of BDE-209 in the Lake Michigan food web.

Electronic supplementary material

Keywords: Polybrominated diphenyl ethers - Decabromodiphenyl ether - Biomagnification - Bioaccumulation - Food web - Lake Michigan

Christopher C. Rimmer[1], Eric K. Miller[2], Kent P. McFarland[1], Robert J. Taylor[3] and Steven D. Faccio[1]. ([1]Vermont Center for Ecostudies, P.O. Box 420, Norwich, VT 05055, USA, [2]Ecosystems Research Group, Ltd., P.O. Box 1227, Norwich, VT 05055, USA, [3]Trace Element Research Laboratory, Department of Veterinary Integrative Biosciences, Texas A&M University, College Station, TX 77843-4458, USA). **Mercury bioaccumulation and trophic transfer in the terrestrial food web of a montane forest. Ecotoxicology, Volume 19(4) (2010): 697-709**

We investigated mercury (Hg) concentrations in a terrestrial food web in high elevation forests in Vermont. Hg concentrations increased from autotrophic organisms to herbivores < detritivores < omnivores < carnivores. Within the carnivores studied, raptors had higher blood Hg concentrations than their songbird prey. The Hg concentration in the blood of the focal study species, Bicknell's thrush (*Catharus bicknelli*), varied over the course of the summer in response to a diet shift related to changing availability of arthropod prey. The Bicknell's thrush food web is more detrital-based (with higher Hg concentrations) in early summer and more foliage-based (with lower Hg concentrations) during late summer. There were significant year effects in different ecosystem compartments indicating a possible connection between atmospheric Hg deposition, detrital-layer Hg concentrations, arthropod Hg concentrations, and passerine blood Hg concentrations.

Keywords: Mercury bioaccumulation - Food web - *Catharus bicknelli* - Montane forests

Yin-Ming Kuo[1, 2], Maria S. SepÃºlveda[1, 3], Trent M. Sutton[5], Hugo G. Ochoa-AcuÃ±a[1, 6], Andrew M. Muir[3, 4], Benjamin Miller[3] and Inez Hua[1, 7]. ([1]School of Civil Engineering, Purdue University, 550 Stadium Mall Dr., West Lafayette, IN 47907, USA, [2]*Present address:* Department of Civil and Environmental Engineering, University of Vermont, 33 Colchester Ave., Burlington, VT 05405, USA, [3]Department of Forestry and Natural Resources, Purdue University, 195 Marsteller St., West Lafayette, IN 47907, USA, [4]*Present address:* Golder Associates Ltd., 9-4905-48th Street, Yellowknife, Northwest Territories, X1A 3S3, Canada, [5]School of Fisheries and Ocean Sciences, University of Alaska Fairbanks, 245 O'Neill Building, Fairbanks, AK 99775, USA, [6]Department of Comparative Pathobiology, Purdue University, 725 Harrison St., West Lafayette, IN 47907, USA, [7]Division of Environmental and Ecological Engineering, Purdue University, 500 Central Dr., West Lafayette, IN 47907, USA). **Bioaccumulation and biotransformation of**

decabromodiphenyl ether and effects on daily growth in juvenile lake whitefish (*Coregonus clupeaformis*). Ecotoxicology, Volume 19(4) (2010): 751-760

Decabromodiphenyl ether (BDE 209) is the main congener in the commonly used commercial flame retardant mixture, "deca-BDE". There is evidence showing that fish can debrominate BDE 209 into potentially more toxic congeners. The objective of this study was to evaluate BDE 209 uptake and its potential effects on juvenile lake whitefish (*Coregonus clupeaformis*). Lake whitefish were fed BDE 209 at four nominal concentrations (control, 0.1, 1, and 2 µg/g-diet) for 30 days. Livers and carcasses were analyzed for 11 polybrominated diphenyl ether (PBDE) congeners (BDE 47, 99, 100, 153, 154, 196, 197, 206, 207, 208, and 209) and daily otolith increment width was measured as an estimate of growth before and after exposure. Four congeners (BDE 206, 207, 208, and 209) were detected in livers and carcasses. Hepatic BDE 209 concentrations in the 1 and 2 µg/g treatments were significantly higher than in the control group (1.25 and 5.80 nmol/g-lipid compared to 0.183 nmol/g-lipid). The concentration of BDE 209 detected in the tissues of the control group resulted from BDE 209 in the base diets. Concentrations of all congeners from the 1 and 2 µg/g groups were higher in livers than carcasses, indicating the liver was the primary organ of BDE 209 accumulation. Compared to the fraction in diets, the molar fraction of BDE 209 was lower in livers and carcasses, whereas the fractions of BDE 206, 207, and 208 were higher. These different distributions of PBDE congeners resulted from differential adsorption and metabolism. One congener, BDE 206, could be a major metabolite from BDE 209 debromination. Otolith increment widths were narrower in fish from the highest diet concentration administered, suggesting BDE 209 may have affected growth rates. In conclusion, this in vivo study with lake whitefish showed that BDE 209 was debrominated into lower PBDE congeners and that exposure to 2 µg/g may have affected fish growth.

Keywords: Lake whitefish - Decabromodiphenyl ether - Debromination - Bioaccumulation - Otoliths - Growth

Jianzhong Chen[1, 2], Jian Dai[3], Haiyang Zhang[1], Chenyi Wang[1], Guoqing Zhou[1], Zhiping Han[1] and Zhili Liu[2]. ([1]School of Life Science, Huzhou University, 313000 Huzhou, People's Republic of China, [2]School of Life Science, Nanjing University, 210093 Nanjing, People's Republic of China, [3]Institute of Food Safety and Detection, Jiangsu Academy of Agricultural Sciences, 210014 Nanjing, People's Republic of China). **Bioaccumulation of microcystin and its oxidative stress in the apple (*Malus pumila*). Ecotoxicology, Volume 19(4) (2010): 796-803**

The bioaccumulation and harmful effects of microcystins (MCs) and the activity of peroxidase (POD) and superoxide dismutase (SOD) were examined in the apple (*Malus pumila*) exposed in vitro with the crude extract of toxic cyanobacterial blooms from Dianchi Lake in southwestern China. The results showed that the growth and proliferation of *M. pumila* shoots in vitro decreased markedly after exposure to microcystins above 0.3 µg/ml. Recovered microcystins determined by enzyme-linked immunosorbent assay (ELISA) in *M. pumila* shoot cultures increased with exposure time and concentration. After 14 days exposure to the concentration of 3 µg/ml microcystins, *M. pumila* shoot cultures accumulated microcystins up to a concentration of 510.23 Â± 141.10 ng MC-LR equiv/g FW (fresh weight), equivalent to an accumulation rate of 36.45 ng/g day. POD activity was significantly increased after 7 days exposure to 3 µg/ml microcystins. After 14 days of exposure, microcystins caused POD to increase significantly at the concentration of 0.3 and 3 µg/ml. The activity of SOD was not affected by microcystins at

concentrations up to 3 µg/ml on 7 days. After 14 days exposure to microcystins, SOD activity increased significantly at the concentration of 0.3 and 3 µg/ml in *M. pumila* shoot cultures.

Keywords: Microcystin - *Malus pumila* - Tissue culture - ELISA - Superoxide dismutase - Peroxidase

James P. Meador[1], Gina M. Ylitalo[2], Frank C. Sommers[1] and Daryle T. Boyd[2]. ([1]Ecotoxicology and Environmental Fish Health Program, Environmental Conservation Division, Northwest Fisheries Science Center, National Marine Fisheries Service, NOAA, 2725 Montlake Boulevard East, Seattle, WA 98112, USA, [2]Environmental Assessment Program, Environmental Conservation Division, Northwest Fisheries Science Center, National Marine Fisheries Service, NOAA, 2725 Montlake Boulevard East, Seattle, WA 98112, USA). **Bioaccumulation of polychlorinated biphenyls in juvenile chinook salmon (*Oncorhynchus tshawytscha*) outmigrating through a contaminated urban estuary: dynamics and application. Ecotoxicology, Volume 19(1) (2010): 141-152**

A field study was conducted to examine bioaccumulation of polychlorinated biphenyls (PCBs) for hatchery-raised and naturally reared (wild) ocean-type juvenile chinook salmon outmigrating through the Lower Duwamish Waterway (LDW), a contaminated urban estuary in Seattle, WA, USA. These results show differences in bioaccumulation of PCBs over time and space in this estuary, which may also occur for any contaminant that is distributed heterogeneously in this system. Highly mobile, outmigrating salmon accumulated ~3–5 times more PCBs on the east side of the LDW than fish on the west side, which is supported by an almost identical difference in mean sediment concentrations. The tPCB concentration data suggest that for most of the spring and early summer, juvenile chinook were likely segregated between the east and west side of the LDW, but may have crossed the channel later in the year as larger fish. Additionally, we used biota-sediment accumulation factors to assess the relative degree of bioaccumulation and explore these factors as potential metrics for predicting adverse sediment concentrations. These results highlight the importance of time and space in sampling design for a highly mobile species in a heterogeneous estuary.

Keywords: PCBs - Bioaccumulation - Salmon - Spatial segregation - Toxicity guideline value

M. Ruiz[1] and J. Velasco[1.] ([1]Department of Ecology and Hydrology, Faculty of Biology, University of Murcia, 30100 Murcia, Spain). **Nutrient Bioaccumulation in *Phragmites australis*: Management Tool for Reduction of Pollution in the Mar Menor. Water, Air, & Soil Pollution, Volume 205(1-4) (2010): 173-185**

We studied nutrient removal by *Phragmites australis* in the Albujón rambla, the main drainage system that discharges into the Mar Menor, a Mediterranean coastal lagoon of high conservation interest, but highly threatened by point and nonpoint pollution derived from tourism and agricultural activities. We measured aerial biomass and N and P concentrations in both aboveground and belowground tissues of common reed during an annual cycle that included two cutting events and two periods of reed growth (one at the end of summer after cutting and another at the beginning of spring, following their natural cycle). The temporal variation of N and P concentrations was related to the phenology of the plant and cutting events. The maximum nutrient concentrations were recorded in young stems in the initial stages of the autumn growing

season (35.86 mg N g^{-1} and 2.38 mg P g^{-1}). The phosphorus dynamics showed evidence of translocation processes related with growth activity, although no evidence of N translocation was found. In November and in summer, when aerial growth ceases because of the hard conditions, the P concentration in rhizomes was higher than in stems, while in spring and in September, the period of maximal growth, the reverse relation was found. The highest total amounts of the two elements in the aboveground biomass (0.54 Tm N ha^{-1} and 0.25 Tm P ha^{-1}) were reached in July, coinciding with the highest biomass (3.72 kg DW m^{-2}), which then decreased to approximately half in August. Nutrient content in the aboveground tissues was highly dependent on the ammonium and nitrate water concentrations. In addition, the N content was inversely related to the C_{org}/N of sediments, while the P content was influenced positively by the phosphorous concentration of the water. Common reed of the Albujón rambla corresponds to the assimilation type, adapted to nutrient-rich habitats, which is characterized by a pronounced external N cycle and P internal reserves. Based on the results obtained, we propose a management plan for common reed to help control eutrophication of the Mar Menor lagoon. This would bring forward reed cutting to the beginning of summer, instead of August, coinciding with the time of maximum aerial biomass, greater nutrient retention, and lower risk of strong precipitation.

Keywords: *Phragmites australis* - Nutrients - Bioaccumulation - Mar Menor - Water pollution

Lingxiangyu Li[a], Zhenlan Xu[a], Jianyang Wu[a] and Guangming Tian[a]. ([a] Department of Environmental Engineering, College of Environmental and Resource Sciences, Zhejiang University, Hangzhou 310029, China). Bioaccumulation of heavy metals in the earthworm *Eisenia fetida* in relation to bioavailable metal concentrations in pig manure. Bioresource Technology, Volume 101(10) (2010): 3430-3436

The study investigated the bioaccumulation of Cu, Zn, Pb and Cd in Eisenia fetida fed on pig manure in greenhouse, and its relationship to bioavailability of these elements in pig manure. Metals in exchangeable, carbonates-bound and Fe–Mn oxides-bound fractions obtained by sequential extraction were considered to be bioavailable. The bioaccumulation factors (BAFs) of E. fetida to heavy metals were Cd (2.749 ± 0.441), Zn (0.594 ± 0.200), Pb (0.274 ± 0.101) and Cu (0.076 ± 0.030). Variations in the Pb and Cd concentrations of E. fetida were best explained by the concentration of exchangeable fraction, while that of Cu was best explained by the concentrations of exchangeable and Fe–Mn oxides-bound fractions. The concentration of Zn in E. fetida depended strongly on the concentrations of exchangeable, carbonates-bound and Fe–Mn oxides-bound fractions. Further nonlinear regression analysis revealed the positive logarithmic relationship between the BAF and the exchangeable metal concentration of pig manure.

Keywords: Pig manure; Heavy metals; Bioavailability; *Eisenia fetida*; Bioaccumulation

Ragini Singh[a], R.D. Tripathi[a], Sanjay Dwivedi[a], Amit Kumar[a], P.K. Trivedi[a] and D. Chakrabarty[a]. ([a] National Botanical Research Institute (Council of Scientific and Industrial Research), Rana Pratap Marg, Lucknow 226 001, Uttar Pradesh, India). Lead bioaccumulation potential of an aquatic macrophyte *Najas indica* are related to antioxidant system. Bioresource Technology, Volume 101(9) (2010): 3025-3032

Plants of *Najas indica* bioaccumulated significantly higher amounts of Pb (3554 μg g^{-1} dw) when, exposed to varying concentrations of $Pb(NO_3)_2$.This also led to increased malondialdehyde (MDA), electrical conductivity (EC) and H_2O_2 content. In response to this, the

activities of antioxidant enzymes such as superoxide dismutase (SOD), ascorbate peroxidase (APX), guaiacol peroxidase (GPX), catalase (CAT) and glutathione reductase (GR) were elevated along with the induction of various molecular antioxidants including GSH, cysteine, ascorbic acid and proline. Further, Pb exposed plants showed significantly increased cysteine synthase and glutathione-*S*-transferase activity. Visible symptoms of toxicity were evident at 50 µM after 4d showing chlorosis and fragmentation of leaves with mucilaginous discharge. It seems that bioaccumulated Pb is efficiently tolerated by *Najas* plants through activation of antioxidant system and thiolic pathways which was evident by the increased biomass up to 10 µM Pb. Therefore, it appears that due to metal tolerance characteristics with high concentration factor these plants can find use in phytoremediation of aquatic system highly contaminated by Pb.

Keywords: Accumulation; Antioxidant enzymes; Glutathione; Lead; *Najas indica*

Bioremediation

Ashutosh Kumar Verma[1], Chandralata Raghukumar[1], Pankaj Verma[2], Yogesh S. Shouche[2] and Chandrakant Govind Naik[1]. ([1]National Institute of Oceanography, Council of Scientific and Industrial Research, Dona Paula, Goa, 403004, India, [2]National Centre for Cell Sciences, Lab-III, Pune University Campus, Ganeshkhind Road, Pune, 411007, India). **Four marine-derived fungi for bioremediation of raw textile mill effluents. Biodegradation, Volume 21(2) (2010): 217-233**

Textile dye effluents pose environmental hazards because of color and toxicity. Bioremediation of these has been widely attempted. However, their widely differing characteristics and high salt contents have required application of different microorganisms and high dilutions. We report here decolorization and detoxification of two raw textile effluents, with extreme variations in their pH and dye composition, used at 20–90% concentrations by each of the four marine-derived fungi. Textile effluent A (TEA) contained an azo dye and had a pH of 8.9 and textile effluent B (TEB) with a pH of 2.5 contained a mixture of eight reactive dyes. The fungi isolated from mangroves and identified by 18S and ITS sequencing corresponded to two ascomycetes and two basidiomycetes. Each of these fungi decolorized TEA by 30–60% and TEB by 33–80% used at 20–90% concentrations and salinity of 15 ppt within 6 days. This was accompanied by two to threefold reduction in toxicity as measured by LC_{50} values against Artemia larvae and 70–80% reduction in chemical oxygen demand and total phenolics. Mass spectrometric scan of effluents after fungal treatment revealed degradation of most of the components. The ascomycetes appeared to remove color primarily by adsorption, whereas laccase played a major role in decolorization by basidiomycetes. A process consisting of a combination of sorption by fungal biomass of an ascomycete and biodegradation by laccase from a basidiomycete was used in two separate steps or simultaneously for bioremediation of these two effluents.

Keywords: Marine fungi - Raw dye-containing effluents - Laccase - Toxicity - Decolorization - Chemical oxygen demand

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Barcelona, Bellaterra, Cerdanyola, 08193 Barcelona,Spain). Optimization and enhancement of soil bioremediation by composting using the experimental design technique. Biodegradation, Volume 21(3) (2010): 345-356

The objective of this study was the application of the experimental design technique to optimize the conditions for the bioremediation of contaminated soil by means of composting. A low-cost material such as compost from the Organic Fraction of Municipal Solid Waste as amendment and pyrene as model pollutant were used. The effect of three factors was considered: pollutant concentration (0.1–2 g/kg), soil:compost mixing ratio (1:0.5–1:2 w/w) and compost stability measured as respiration index (0.78, 2.69 and 4.52 mg O_2 g^{-1} Organic Matter h^{-1}). Stable compost permitted to achieve an almost complete degradation of pyrene in a short time (10 days). Results indicated that compost stability is a key parameter to optimize PAHs biodegradation. A factor analysis indicated that the optimal conditions for bioremediation after 10, 20 and 30 days of process were (1.4, 0.78, 1:1.4), (1.4, 2.18. 1:1.3) and (1.3, 2.18, 1:1.3) for concentration (g/kg), compost stability (mg O_2 g^{-1} Organic Matter h^{-1}) and soil:compost mixing ratio, respectively.

Keywords: Soil bioremediation - Compost stability - Experimental design - Pyrene - Municipal solid waste

Prabhat Kumar Rai[1] . ([1]Environmental Sciences (FEBES), Mizoram University, Tanhril, P.B. 190, Aizawl, Mizoram, India, 796009). Phytoremediation of heavy metals in a tropical impoundment of industrial region. Environmental Monitoring and Assessment, Volume 165(1-4) (2010): 529-537

Aquatic pollution pose a serious challenge to the scientific community worldwide, since lakes or reservoirs find multifarious use and most often their water is used for drinking, bathing, irrigation, and aquaculture. Nine metals and several physicochemical parameters, from four sampling sites in a tropical lake receiving the discharges from a thermal power plant, a coal mine, and a chlor-alkali industry, were studied from 2004 to 2005. Pertaining to metal pollution, the site most polluted with heavy metals was Belwadah, i.e., waters and sediments had the highest concentration of all the metals examined. The reference site was characterized by the presence of low concentrations of metals in waters and sediments. Following the water quality monitoring, 2-month field phytoremediation experiments were conducted using large enclosures at the discharge point of different polluted sites of the lake. During field phytoremediation experiments using aquatic macrophytes, marked percentage reduction in metals concentrations were recorded. The percentage decrease for different metals was in the range of 25% to 67.90% at Belwadah (with *Eichhornia crassipes* and *Lemna minor*), 25% to 77.14% at Dongia nala (with *E. crassipes, L. minor and Azolla pinnata*), and 25% to 71.42% at Ash pond site of G.B. Pant Sagar (with *L. minor and A. pinnata*). Preliminary studies of polluted sites are useful for improved microcosm design and for the systematic extrapolation of information from experimental ecosystems to natural ecosystems.

Keywords: Heavy metals - Water pollution - Phytoremediation - *Eichhornia* - Coal mines

Grażyna A. Płaza[1], Grzegorz Nałęcz-Jawecki[2], Onruthai Pinyakong[3], Paul Illmer[4] and Rosa Margesin[4] . ([1]Department of Environmental Microbiology, Institute for Ecology of Industrial Areas, 40-844 Katowice, Poland, [2]Department of Environmental Health Sciences, Medical University of Warsaw, 02-097 Warszawa, Poland, [3]Department of

Microbiology, Faculty of Science, Chulalongkorn University, Phyathai Road, Patumwan, Bangkok, 10330, Thailand, [4]Institute of Microbiology, University of Innsbruck, Technikerstrasse 25, 6020 Innsbruck, Austria). Ecotoxicological and microbiological characterization of soils from heavy-metal- and hydrocarbon-contaminated sites. Environmental Monitoring and Assessment, Volume 163(1-4) (2010): 477-488

The aims of this study were to characterize soils from industrial sites by combining physicochemical, microbiological, and ecotoxicological parameters and to assess the suitability of these assays for evaluation of contaminated sites and ecological risk assessment. The soil samples were taken from long-term contaminated sites containing high amounts of heavy metals (sites 1 and 2) or petroleum hydrocarbons (site 3) located in the upper Silesia Industrial Region in southern Poland. Due to soil heterogeneity, large differences between all investigated parameters were measured. Microbiological properties revealed the presence of high numbers of viable hetrotrophic microorganisms. Soil enzyme activities were considerably reduced or could not be detected in contaminated soils. Activities involved in N turnover (N mineralization and nitrification) were significantly ($P<0.05$) higher in samples from the metal-contaminated sites than in samples from the hydrocarbon-contaminated site, whereas the opposite was observed for phosphatase activity. The Microtox test system appeared to be the most appropriate to detect toxicity and significant differences in toxicity between the three sites. The Ostracodtoxkit test was the most appropriate test system to detect toxicity in the hydrocarbon-contaminated soil samples. Correlation analysis between principal components (obtained from factor analysis) determined for physicochemical, microbiological, and ecotoxicological soil properties demonstrated the impact of total and water-extractable contents of heavy metals on toxicity.

Keywords: Soil pollution - Heavy metals - Hydrocarbons - Microbial activity - Ecotoxicity

Ajay K. Manna[a], Mou Sen[a], Andrew R. Martin[b] and Parimal Pal[a]. ([a] Environment and Membrane Technology Laboratory, Department of Chemical Engineering, National Institute of Technology, Durgapur 713209 India, [b] Department of Energy Technology, The Royal Institute of Technology (KTH), Stockholm, Sweden). Removal of arsenic from contaminated groundwater by solar-driven membrane distillation. Environmental Pollution, Volume 158(3) (2010): 805-811

Experimental investigations were carried out on removal of arsenic from contaminated groundwater by employing a new flat-sheet cross flow membrane module fitted with a hydrophobic polyvinylidenefluoride (PVDF) microfiltration membrane. The new design of the solar-driven membrane module in direct contact membrane distillation (DCMD) configuration successfully produced almost 100 per cent arsenic-free water from contaminated groundwater in a largely fouling-free operation while permitting high fluxes under reduced temperature polarization. For a feed flow rate of 0.120 m^3/h, the 0.13 µm PVDF membrane yielded a high flux of 74 $kg/(m^2\ h)$ at a feed water temperature of 40 °C and, 95 $kg/m^2\ h$ at a feed water temperature of 60 °C. The encouraging results show that the design could be effectively exploited in the vast arsenic-affected rural areas of South-East Asian countries blessed with abundant sunlight particularly during the critical dry season.

Solar-driven membrane distillation has the potential of removing arsenic from contaminated groundwater.

Keywords: Membrane distillation; Solar energy; Arsenic separation; Cross flow module; Hydrophobic membrane; Microfiltration

Stefanie Knauert[a], Heinz Singer[b], Juliane Hollender[b] and Katja Knauer[c]. ([a]University of Basel, Department of Environmental Sciences, Hebelstrasse 1, 4056 Basel, Switzerland, [b]Eawag: Swiss Federal Institute of Aquatic Science and Technology, Department of Environmental Chemistry, Überlandstrasse 133, 8600 Dübendorf, Switzerland, [c]University of Basel, Program Sustainability Research, Klingelbergstr. 50, 4056 Basel, Switzerland). **Phytotoxicity of atrazine, isoproturon, and diuron to submersed macrophytes in outdoor mesocosms. Environmental Pollution, Volume 158(1) (2010): 167-174**

The submersed macrophytes *Elodea canadensis*, *Myriophyllum spicatum* and *Potamogeton lucens* were constantly exposed over a five-week period to environmentally relevant concentrations of atrazine, isoproturon, diuron, and their mixture in outdoor mesocosms. Effects were evaluated investigating photosynthetic efficiency (PE) of the three macrophytes and growth of *M. spicatum* and *E. canadensis*. Adverse effects on PE were observed on days 2 and 5 after application. *M. spicatum* was found to be the more sensitive macrophyte. *E. canadensis* and *P. lucens* were less sensitive to atrazine, diuron and the mixture and insensitive to isoproturon. PE of *M. spicatum* was similarly affected by the single herbicides and the mixture demonstrating concentration addition. Growth of *E. canadensis* and *M. spicatum* was not reduced indicating that herbicide exposure did not impair plant development. Although PE measurements turned out to be a sensitive method to monitor PSII herbicides, plant growth remains the more relevant ecological endpoint in risk assessment.

Short-term effects on photosynthesis did not result in growth reduction of submerse macrophytes exposed to PSII inhibitors.

Keywords: Photosystem II inhibitors; Species sensitivity; In vivo chlorophyll fluorescence; Growth; Mixtures; Aquatic plants

N. Suchkova[a, 1], E. Darakas[b, 2,] and J. Ganoulis[b, 2,]. ([a] Department of Urban Environmental Engineering and Management, National Academy of Municipal Economy, Revolutsii Str.12, Kharkiv 61002, Ukraine, [b] Department of Civil Engineering, Aristotle University of Thessaloniki, Gr-54124 Thessaloniki, Greece). **Phytoremediation as a prospective method for rehabilitation of areas contaminated by long-term sewage sludge storage: A Ukrainian–Greek case study. Ecological Engineering, Volume 36(4) (2010): 373-378**

Soil contamination by heavy metals could be caused by long-term storage of sewage sludge on the territory of most municipal wastewater treatment plants (WWTPs) worldwide. Different methods to deal with heavy metal pollution and rehabilitation can be applied, but they are costly. Phytoremediation is a method using plants in order to extract, sequester and/or detoxify pollutants such as heavy metals. Phytotechnologies are more advantageous economically, than other in situ and ex situ remedial approaches (they estimated to be at least 40% less costly) (ITRC, 2001).

In this work the suitability of several plant species for phytoremediation under natural conditions was studied. *Brassica napus*, *Medicago sativa*, *Zea mays*, *Triticum aestivum* and *Hordeum vulgare* were grown in pots with sewage sludge from “Bezludivka” WWTP in Kharkiv, Ukraine and from Sindos WWTP in Thessaloniki, Greece.

Plants in the experimental series were compared to those in the control samples (the same species grown in compost). In experimental series, shoot growth was less reduced in *T. aestivum* and *H. vulgare* than in the other plant species studied. *M. sativa* had the lowest germination rate. Generally *B. napus* and *M. sativa*, giving less biomass production than *Z. mays* and *T. aestivum*, were characterized by higher ability to accumulate heavy metals (Cd, Cu, Ni, Pb, Zn, Cr, As and Hg).

Keywords: Phytoremediation; Soil contamination; Sewage sludge; Heavy metals; Germination

Xiao-Zhang Yu[1], Xiao-Ying Peng[1] and Li-Qun Xing[1]. ([1]Department of Environmental Sciences & Engineering, Hunan Agricultural University, 41028 Changsha, People's Republic of China). **Effect of temperature on phytoextraction of hexavalent and trivalent chromium by hybrid willows. Ecotoxicology, Volume 19(1) (2010): 61-68**

The removal of hexavalent and trivalent chromium from hydroponic solution by plants to changes in temperature was investigated. Pre-rooted hybrid willows (*Salix matsudana Koidz* Ã— *alba* L.) were exposed to a nutrient solution spiked with potassium chromate (K_2CrO_4) or chromium chloride ($CrCl_3$) for 4 days. Ten different temperatures were tested ranging from 11 to 32Â°C. Total Cr in solutions and in plant materials were all analyzed quantitatively. The results revealed that large amounts of the applied Cr were removed from the hydroponic solution in the presence of the plants. Significantly faster removal of Cr(III) than Cr(VI) was achieved by hybrid willows from the hydroponic solutions at all temperatures ($P < 0.01$). The removal rates of both chemical forms of Cr by plants increased linearly with the increase of temperatures. The highest removal rate of Cr(VI) was found at 32Â°C with a value of 1.99 µg Cr/g day, whereas the highest value of Cr(III) was 3.55 µg Cr/g day at the same temperature. Roots were the main sink for Cr accumulation in plants at all temperatures. Translocation of both chemical forms of Cr from roots to lower stems was only found at temperatures ≥24Â°C. The temperature coefficient values (Q_{10}) were 2.41 and 1.42 for Cr(VI) and Cr(III), respectively, indicating that the removal of Cr(VI) by hybrid willows was much more susceptible to changes in temperature than that of Cr(III). This information suggests that changes in temperature have a substantial influence on the uptake and accumulation of both chemical forms of Cr by plants.

Keywords: Accumulation - Chromium - Phytoremediation - Removal - Temperature - Willows

Fatih Duman[1], Fatma Ozturk[1] and Zeki Aydin[2]. ([1]Department of Biology, Faculty of Arts and Sciences, Erciyes University, 38039 Kayseri, Turkey, [2]Department of Chemistry, Faculty of Arts and Sciences, Mustafa Kemal University, 31040 Hatay, Turkey). **Biological responses of duckweed (*Lemna minor* L.) exposed to the inorganic arsenic species As(III) and As(V): effects of concentration and duration of exposure. Ecotoxicology, Volume 19(5) (2010): 983-993**

The accumulation of arsenic (As) and physiological responses of *Lemna minor* L. under different concentration (0, 1, 4, 16 and 64 µM) and duration (1, 2, 4 and 6 days) of two species As, $NaAsO_2$ and Na_2HAsO_4Â·$7H_2O$, were studied in hydroponics. The accumulation of both As species depended on As concentration and exposure duration. The highest accumulation of As was found as 17408 and 8674 µg g^{-1}, for plants exposed to 64 µM of As(III) and As(V),

respectively, after 6 days. Two-way ANOVA analyses indicated that, for plants exposed to arsenite (As(III)), exposure duration had a greater effect than concentration on As accumulation. Conversely, exposure concentration had a greater effect on As accumulation in plants exposed to arsenate (As(V)). Arsenic exposure levels, approaching 16 µM for As(III) and 64 µM for As(V), did not significantly affect EC values. Beyond these exposure concentrations, EC values increased in a manner that depended on duration. Significant effect of As(III) on lipid peroxidation was observed at 1 µM application whereas, this effect started to be significant after an exposure to 16 µM As(V). For both As(III) and As(V), photosynthetic pigment levels slightly increased for the first day with respect to the control, followed by a gradual decline at higher concentrations and durations. An increase in protein content and enzyme activity was observed at moderate exposure conditions, followed by a decrease. Significant positive correlations were determined between accumulated As and ion leakage and lipid peroxidation. Negative correlations were found between accumulated As and total chlorophyll and protein content. Our results suggested that exposure duration and concentration had a strong synergetic effect on antioxidant enzyme activity. The findings of the present study may be useful when this plant is used as a phytoremediator in arsenic-polluted water.

Keywords: Arsenite - Arsenate - *Lemna minor* - Bioaccumulation - Exposure - Duration

Md. Abul Kashem[1], Bal Ram Singh[2], Hiroshi Kubota[3], Reiko Sugawara[3], Nobuyuki Kitajima[3], Toshihito Kondo[3] and Shigenao Kawai[1]. ([1]Laboratory of Plant Physiology and Nutrition, Iwate University, Ueda 3-18-8, Morioka 020-8550, Japan, [2]Department of Plant and Environmental Sciences, Norwegian University of Life Sciences, P.O. Box 5003, 1432 Aas, Norway, [3]Technology Development Division, Fujita Corporation, 2025-1 Ono, Atsugi 243-0125, Japan). **Zinc tolerance and uptake by *Arabidopsis halleri ssp. gemmifera* grown in nutrient solution. Environmental Science and Pollution Research, Volume 17(5) (2010): 1174-1176**

Background, aim, and scope

Zinc is an essential micronutrient element but its concentrations found in contaminated soils frequently exceed those required by the plant and soil organisms, and thus create danger to animal and human health. Phytoremediation is a technique, often employed in remediation of contaminated soils, which aims to remove heavy metals or other contaminants from soils or waters using plants. *Arabidopsis* (*A.*) *halleri ssp. gemmifera* is a plant recently found to be grown vigorously in heavy metal contaminated areas of Japan and it contained remarkably high amount of heavy metals in its shoots. However, the magnitude of Zn accumulation and tolerance in *A. halleri ssp. gemmifera* need to be investigated for its use as a phytoremediation plant.

Materials and methods

A. halleri ssp. gemmifera was grown for 3 weeks into half-strength nutrient solution with Zn (as $ZnSO_4$) levels ranging from 0.2 to 2,000 ÂµM. The harvested plants were separated into shoots and roots, dried in the oven, and ground. The plant tissue was digested with nitric-perchloric acid, and the Zn concentration in the digested solution was measured by atomic absorption spectrophotometer.

Results and discussion

The results showed no reduction in shoot and root dry weight when plants were grown at 0.2 to 2,000 ÂµM Zn in the solution. The highest Zn concentration measured in the shoots was 26,400 mg kg^{-1} at 1,000 ÂµM Zn, while in the roots, it was 71,000 mg kg^{-1} at 2,000 ÂµM Zn treatment. Similar to the Zn concentration in plant parts, maximum Zn accumulation of 62 mg $plant^{-1}$ in the shoots and 22 mg $plant^{-1}$ in the roots was obtained at 1,000 and 2,000 ÂµM Zn in the solution. The percentage of Zn translocation in shoot varied from 69% to 90% of the total Zn, indicating that the shoot was the major sink of Zn accumulation in this plant.

Conclusions

The results of this study indicate that the growth of *A. halleri ssp. gemmifera* was not affected by the Zn level of up to 2,000 ÂµM in the nutrient solution. The concentration of Zn found in shoot indicated that *A. halleri ssp. gemmifera* has an extraordinary ability to tolerate and accumulate Zn and hence a good candidate for the phytoremediation of Zn-polluted soil.

Recommendations and outlook

Based on the results presented in this study and earlier hydroponics, and field study, *A. halleri ssp. gemmifera* seems to be a potential heavy metals hyperaccumulator, and could be recommended to use for phytoremediation of Cd- and Zn-contaminated soils.

Keywords: Hyperaccumulator - Metal tolerance - Nutrient solution - Phytoremediation - Zinc

Metka Udovic[1] and Domen Lestan[1]. ([1]Biotechnical Faculty, Agronomy Department, Centre for Soil and Environmental Science, University of Ljubljana, Jamnikarjeva 101, 1000 Ljubljana, Slovenia). **Fractionation and bioavailability of Cu in soil remediated by EDTA leaching and processed by earthworms (*Lumbricus terrestris* L.). Environmental Science and Pollution Research, Volume 17(3) (2010): 561-570**

Background, aim, and scope

Soil remediation with ethylenediamine tetraacetic acid (EDTA) leaching is capable of removing only part of the total metal concentration in the soil, mostly the labile, bioavailable metal species (metal bioavailability stripping). However, reintroduction of remediated soil in the environment exposes the soil to various environmental factors, which could potentially shift nonlabile residual metals back to labile bioavailable forms. We studied the effect of autochthonous earthworm species as model biotic environmental factor on the fractionation and bioavailability of Cu residual in soil after remediation.

Materials and methods

We used soil from a 50-year-old vineyard regularly managed and treated with $CuSO_4 \cdot 5H_2O$ (Bordeaux mixture) as fungicide. Soil containing 400 mg kg^{-1} of Cu was leached with total 15 mmol kg^{-1} EDTA. Remediated and nonremediated soil was processed by fully clitellated adult specimens of *Lumbricus terrestris* L., a prevailing autochthonous soil earthworm species.

Cu fractionation, phytoavailability, and oral-bioavailability in processed and nonprocessed soil were determined using six-step sequential extraction, extraction with diethylenediamine pentaacetic acid, and in vitro physiologically based extraction test, respectively.

Results

EDTA leaching removed 41% of the pseudototal Cu, mostly from the soil Fe- and Mn-oxides, carbonates, and organic matter. A 2.7-fold decrease in Cu phytoavailability and a 4.4- and 2.8-fold decrease in Cu oral-bioavailability in the stomach and small intestine fractions, respectively, were achieved after remediation. In nonremediated soil, earthworms increased the share of nonlabile Cu in residual soil fraction, while in remediated soil they increased the share of Cu bound to carbonates. A statistically significant 1.1- and 1.7-fold increase in Cu phytoavailability and intestinal oral-bioavailability, respectively, was observed in earthworm processed remediated soil.

Discussion

Cu occurs in various soil "pools" of different solubilities with different chemical characteristics and consequently different functions. By removing the labile part of the metals from the soil during remediation, we disrupt the chemical equilibrium; the nonlabile residual metals left in soil after remediation might become more labile in time in tendency to re-establish that equilibrium. Earthworms alter the physical and chemical properties of soil affecting consequently the fractionation of metals. The increase in earthworm's gut pH due to the excretion of ammonia and/or calcium carbonate into the intestine could lead to the transbounding of metals into the carbonate fraction. However, their activity in remediated soil increased Cu phytoavailability and intestinal oral-bioavailability, and it would, therefore, be improper to generalize the influence of earthworms on metal availability in soil.

Conclusions

The results presented here show that residual Cu in remediated soil is affected by environmental factors such as earthworms, which should be considered in evaluating the effect of Cu polluted soil remediation.

Recommendations and perspectives

Information on the behavior of residual metals in soil after its remediation is surprisingly scarce. The development of new effective remediation techniques should imply also the evaluation of postremediation effects on remediated soil. The results presented in this work indicate a possible tool for assessing the effect of biotic environmental factors on residual metals left in soil after its remediation.

Keywords: Cu - Cu bioavailability - Cu phytoavailability - Ethylenediamine tetraacetic acid (EDTA) - *Lumbricus terrestris* - Soil leaching - Soil remediation

Nitu Sood[1], Sonali Patle[1] and Banwari Lal[2]. ([1]TERI University, India Habitat Centre, Lodhi Road, New Delhi, India, 110 003, [2]The Energy and Resources Institute, India Habitat Centre, Lodhi Road, New Delhi, India, 110 003). Bioremediation of acidic oily sludge-contaminated soil by the novel yeast strain *Candida digboiensis* TERI ASN6. Environmental Science and Pollution Research, Volume 17(3) (2010): 603-610

Background, aim, and scope

Primitive wax refining techniques had resulted in almost 50,000 tonnes of acidic oily sludge (pH 1–3) being accumulated inside the Digboi refinery premises in Assam state, northeast India. A novel yeast species *Candida digboiensis* TERI ASN6 was obtained that could degrade the acidic petroleum hydrocarbons at pH 3 under laboratory conditions. The aim of this study was to evaluate the degradation potential of this strain under laboratory and field conditions.

Materials and methods

The ability of TERI ASN6 to degrade the hydrocarbons found in the acidic oily sludge was established by gravimetry and gas chromatography–mass spectroscopy. Following this, a feasibility study was done, on site, to study various treatments for the remediation of the acidic sludge. Among the treatments, the application of *C. digboiensis* TERI ASN6 with nutrients showed the highest degradation of the acidic oily sludge. This treatment was then selected for the full-scale bioremediation study conducted on site, inside the refinery premises.

Results

The novel yeast strain TERI ASN6 could degrade 40 mg of eicosane in 50 ml of minimal salts medium in 10 days and 72% of heneicosane in 192 h at pH 3. The degradation of alkanes yielded monocarboxylic acid intermediates while the polycyclic aromatic hydrocarbon pyrene found in the acidic oily sludge yielded the oxygenated intermediate pyrenol. In the feasibility study, the application of TERI ASN6 with nutrients showed a reduction of solvent extractable total petroleum hydrocarbon (TPH) from 160 to 28.81 g kg^{-1} soil as compared to a TPH reduction from 183.85 to 151.10 g kg^{-1} soil in the untreated control in 135 days. The full-scale bioremediation study in a 3,280-m^2 area in the refinery showed a reduction of TPH from 184.06 to 7.96 g kg^{-1} soil in 175 days.

Discussion

Degradation of petroleum hydrocarbons by microbes is a well-known phenomenon, but most microbes are unable to withstand the low pH conditions found in Digboi refinery. The strain *C. digboiensis* could efficiently degrade the acidic oily sludge on site because of its robust nature, probably acquired by prolonged exposure to the contaminants.

Conclusions

This study establishes the potential of novel yeast strain to bioremediate hydrocarbons at low pH under field conditions.

Recommendations and perspectives

Acidic oily sludge is a potential environmental hazard. The components of the oily sludge are toxic and carcinogenic, and the acidity of the sludge further increases this problem. These results establish that the novel yeast strain *C. digboiensis* was able to degrade hydrocarbons at low pH and can therefore be used for bioremediating soils that have been contaminated by acidic hydrocarbon wastes generated by other methods as well.

Keywords: Acidic oily sludge - Bioremediation - *Candida digboiensis* - Contaminated soil - Degradation - Total petroleum hydrocarbon

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Phytotoxicity tests of solid wastes and contaminated soils in the Czech Republic. Environmental Science and Pollution Research, Volume 17(3) (2010): 611-623

Background, aim, and scope

The purpose of this study was to compare the suitability of different phytotoxicity testing procedures for the evaluation of toxicity associated with both soil contamination and solid wastes, both of which can be of environmental risk to plants. Ten different representative types of contaminated soils and solid waste samples were chosen from the Czech Republic.

Materials and methods

Both solid-phase and aquatic toxicity testing procedures on mono- and dicotyledonous plants were performed using *Lactuca sativa* L., *Sinapis alba* L., *Hordeum vulgare* L., *Triticum aestivum* L., *Lemna minor* L., and the chlorococcal algae *Desmodesmus subspicatus* (syn. *Scenedesmus subspicatus*), strain BRINKMANN 1953/SAG 86.81. An innovative classification scheme, using the intensity of toxic effects upon the plants, is presented in the study. Detailed chemical characterizations of both solid samples and their aquatic elutriates were carried out, using the appropriate ISO guidelines. In the solid samples, all the congeners of polychlorinated biphenyls were analyzed, together with 16 U.S. EPA polyaromatic hydrocarbons, the aggregate of C10–C40 hydrocarbons, total organic carbon, extractable organic halogens, as well as the majority of the environmentally toxic metals. In the aquatic elutriates, parameters analyzed were pH, conductivity, dissolved organic content, phenol index, main anions, and the majority of the environmentally relevant metals.

Results

Eight out of ten samples tested expressed phytotoxic properties on tested organisms. Only three of the samples were toxic to both aquatic and terrestrial organisms in the tests. This demonstrates how different substances present in different samples can express different types of toxic effects, resulting in the illogical substituting terrestrial bioassays with aquatic ones.

Discussion

Based upon our experience, we propose the following battery of bioassays for use in the characterization of toxic properties of solid wastes and contaminated soils: Aquatic ecosystems were tested by the algae *D. subspicatus* and plant *L. minor*; and the terrestrial ecosystems were tested by the dicotyledonous *L. sativa* and monocotyledonous *H. vulgare*. This proposed new battery of bioassays for the detection of phytotoxicity of both solid wastes and contaminated soils has higher sensitivity (as well as greater ecological relevance) compared to the battery of bioassays currently used in the Czech Republic.

Conclusions

The tests currently used for regulatory purposes in the Czech Republic are phytotoxicity tests of elutriates, using *S. alba* and *D. subspicatus*, which have been found insufficiently sensitive to the range of different pollutants present in contaminated soils and/or solid wastes. If only aquatic bioassays are used for the toxicity testing, it is possible that the toxic effects of substances (poorly or totally) insoluble in water might be underestimated. The new proposed system of toxicity classification has proven to be both practical and sensitive.

Recommendations and perspectives

This recommended alternative battery of phytotoxicity tests includes both aquatic tests of waste elutriates (with the algae *D. subspicatus* along with the aquatic plant *L. minor*), in addition to tests of the terrestrial solid samples (with the dicotyledonous *L. sativa* and the monocotyledonous *H. vulgare*). This battery of bioassays is sufficiently sensitive, representing a majority of types of aquatic and terrestrial plants.

Keywords: Ecotoxicity - Phytotoxicity - Soil contamination - Solid waste - Toxicity - Toxicity testing - Waste classification - Waste toxicity

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Phytoremediation to remove nutrients and improve eutrophic stormwaters using water lettuce (*Pistia stratiotes* L.). Environmental Science and Pollution Research, Volume 17(1) (2010): 84-96

Background, aim, and scope Water quality impairment by nutrient enrichment from agricultural activities has been a concern worldwide. Phytoremediation technology using aquatic plants in constructed wetlands and stormwater detention ponds is increasingly applied to remediate eutrophic waters. The objectives of this study were to evaluate the effectiveness and potential of water lettuce (*Pistia stratiotes* L.) in removing nutrients including nitrogen (N) and phosphorus (P) from stormwater in the constructed water detention systems before it is discharged into the

St. Lucie Estuary, an important surface water system in Florida, using phytoremediation technologies.

Materials and methods In this study, water lettuce (*P. stratiotes*) was planted in the treatment plots of two stormwater detention ponds (East and West Ponds) in 2005–2007 and water samples from both treatment and control plots were weekly collected and analyzed for water quality properties including pH, electrical conductivity, turbidity, suspended solids, and nutrients (N and P). Optimum plant density was maintained and plant samples were collected monthly and analyzed for nutrient contents.

Results Water quality in both ponds was improved, as evidenced by decreases in water turbidity, suspended solids, and nutrient concentrations. Water turbidity was decreased by more than 60%. Inorganic N (NH_4^+ and NO_3^-) concentrations in treatment plots were more than 50% lower than those in control plots (without plant). Reductions in both PO_4^{3-} and total P were approximately 14–31%, as compared to the control plots. Water lettuce contained average N and P concentrations of 17 and 3.0 g kg^{-1}, respectively, and removed 190–329 kg N ha^{-1} and 25–34 kg P ha^{-1} annually.

Discussion Many aquatic plants have been used to remove nutrients from eutrophic waters but water lettuce proved superior to most other plants in nutrient removal efficiency, owing to its rapid growth and high biomass yield potential. However, the growth and nutrient removal potential are affected by many factors such as temperature, water salinity, and physiological limitations of the plant. Low temperature, high concentration of salts, and low concentration of nutrients may reduce the performance of this plant in removing nutrients.

Conclusions The results from this study indicate that water lettuce has a great potential in removing N and P from eutrophic stormwaters and improving other water quality properties.

Kheirghadam Enayatzamir[1, 2], Hossein A. Alikhani[2], Bagher Yakhchali[3], Fatemeh Tabandeh[3] and Susana RodrÃguez-Couto[1, 4, 5]. ([1]Department of Chemical Engineering, Rovira i Virgili University, 43007 Tarragona, Spain, [2]Department of Soil Science Engineering, Faculty of Soil and Water Engineering, University College of Agriculture and Natural Resources, Tehran University, Karaj, Iran, [3]Department of Bioprocess Engineering, National Institute for Genetic Engineering and Biotechnology, P.O. Box 14155, 6343 Tehran, Iran, [4]CEIT, Section of Environmental Engineering, Paseo Manuel de LardizÃ¡bal 15, 20018 San Sebastian, Spain, [5]Ikerbasque, Basque Foundation for Science, Alameda de Urquijo 36, 48011 Bilbao, Spain). Decolouration of azo dyes by *Phanerochaete chrysosporium* immobilised into alginate beads. Environmental Science and Pollution Research, Volume 17(1) (2010): 145-153

Background, aim and scope Because of high discharged volumes and effluent composition, wastewater from the textile industry can be considered as the most polluting amongst all industrial sectors, thus greatly requiring appropriate treatment technologies. Although some abiotic methods for the reduction of several dyes exist, these require highly expensive catalysts and reagents. Biotechnological approaches were proven to be potentially effective in the treatment of this pollution source in an eco-efficient manner. The white-rot fungi are, so far, the most efficient microorganisms in degrading synthetic dyes. This white-rot fungi's property is due to the production of extracellular lignin-modifying enzymes, which are able to degrade a wide range of xenobiotic compounds because of their low substrate specificity. In this paper, we

studied the ability of the white-rot fungus *Phanerochaete chrysosporium* immobilised into Ca-alginate beads to decolourise different recalcitrant azo dyes such as Direct Violet 51 (DV), Reactive Black 5 (RB), Ponceau Xylidine (PX) and Bismark Brown R (BB) in successive batch cultures. To the best of our knowledge, this is the first study on the immobilisation of *P. chrysosporium* into Ca-alginate beads for its application in dye decolouration.

Materials and methods *P. chrysosporium* was immobilised into Ca-alginate beads using a method of gel recoating to minimise cellular leaking. The immobilised fungus was transferred to 250-ml Erlenmeyer flasks containing 50 ml of growth medium and incubated on an orbital shaker at 150 rpm and 30Â°C for 7 days. The ratio of beads/medium used was 10% (*w*/*v*). The dyes were added into the culture flasks when MnP production started (50 U l^{-1}), which corresponded with the seventh cultivation day. MnP activity and dye decolouration were measured spectrophotometrically.

Results The dyes DV, RB and PX were almost totally decolourised at the end of each batch during the course of three successive batches. However, the dye BB was more resistant to decolouration and it was not completely decolourised (86.7% in 144 h). Further, the beads were kept in sterilised calcium chloride (2 g l^{-1}) for 3 weeks at 4Â°C. After these three storage weeks, the immobilised *P. chrysosporium* was again efficiently reused for azo dye decolouration during two successive batches, decolouration being more effective even for BB. Also, the in vitro decolouration of the aforementioned azo dyes by crude MnP from *P. chrysosporium* was performed. The decolouration levels obtained were lower than those attained with the whole cultures especially for RB and BB dyes, in spite of the fact that dye concentrations used were considerable lower.

Discussion The good performance of the immobilisation system was likely due to the gel re-coating method utilised to prepare the alginate beads which not only maintained the beads integrity but also avoided cellular leaking. The lower decolouration percentages obtained by the enzyme indicates that the mycelial biomass may supply other intracellular or mycelial-bound enzymes, or other compounds that favour dye decolouration.

Conclusions Immobilised *P. chrysosporium* efficiently decolourised different types of azo dyes. In this decolouration process, the MnP secreted by the fungus played the main role whilst adsorption was found to be negligible except for the dye BB.

Recommendations and perspectives Efforts should be made to scale up and apply fungal decolouration techniques to real industrial dye-containing wastewater. Further, detailed characterisation of the intermediates and metabolites produced during biodegradation must be done to ensure the safety of the decolourised wastewater.

Keywords: Alginate beads - Azo dyes - Decolouration - Immobilisation - Ligninolytic enzymes - MnP - *Phanerochaete chrysosporium* - White-rot fungi

Qayyum Husain[1] (Department of Biochemistry, Faculty of Life Sciences, Aligarh Muslim University, Aligarh, 202002, India). Peroxidase mediated decolorization and remediation of wastewater containing industrial dyes: a review. Reviews in Environmental Science and Biotechnology, Volume 9 (2) (2010): 117-140

In this article an effort has been made to review literature based on the role of peroxidases in the treatment and decolorization of a wide spectrum aromatic dyes from polluted water. Peroxidases can catalyze degradation/transformation of aromatic dyes either by precipitation or by opening the aromatic ring structure. Peroxidases from plant sources; horseradish, turnip, tomato, soybean, bitter gourd, white radish and Saccharum uvarum and microbial sources; lignin peroxidases, manganese peroxidases, vanadium haloperoxidases, versatile peroxidases, dye decolorizing peroxidases have been employed for the remediation of commercial dyes. Soluble and immobilized peroxidases have been successfully exploited in batch as well as in continuous processes for the treatment of synthetic dyes with complex aromatic molecular structures present in industrial effluents at large scale. However, recalcitrant dyes were also decolorized by the action of peroxidases in the presence of redox mediators.

Keywords: Peroxidases - Decolorization - Remediation - Aromatic dyes - Industrial effluent - Color

Pooja Singh[1], Othman Sulaiman[1], Rokiah Hashim[1], P. F. Rupani[2] and Leh Cheu Peng[1]. ([1]Bioresource, Paper and Coatings Technology, School of Industrial Technology, Universiti Sains Malaysia, 11800 Pulau Pinang, Malaysia, [2] Environmental Technology Division, School of Industrial Technology, Universiti Sains Malaysia, 11800 Pulau Pinang, Malaysia). Biopulping of lignocellulosic material using different fungal species: a review. Reviews in Environmental Science and Biotechnology, Volume 9 (2) (2010): 141-151

Biopulping can be an alternative to the traditional methods of pulping**.** Biopulping use fungi that are known to be able to degrade wood as well as lignin constituent of wood. Amongst these white rot fungi are the most proficient biodegrader. The fungus is non sporulating and is a selective lignin degrader. It colonizes either on living or dead wood and decomposes all wood polymers including lignin and extractives making it to be extremely potential to be used in biopulping. The process of biopulping reduces the utilization of chemical in pulping industry and help in decreasing the environmental hazard caused by normal pulping. The present review deals with diverse aspects of biopulping and their ecological as well as economic significances.

Keywords: Biopulping - Lignocellulosic material - Fungal species - Lignin - Cellulose - Hemicellulose

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During recent years there has been an increasing interest in the bacterial communities occurring in unusual, often extreme, environments. On serpentine outcrops around the world, a high

diversity of plant species showing the peculiar features of metal hyperaccumulation is present. These metal hyperaccumulators have received much attention for their potential biotechnological exploitation in phytoremediation processes, but also as unusual, extreme habitats for the associated bacterial flora, which could reveal novel details concerning bacterial adaptation. This paper will briefly focus on the research topics that have been addressed to date on bacteria associated with serpentine plants and aims to provide a state of the art and to present possible future directions for research which could lead to new insights on microbial adaptation and evolution, and potentially applied in technologies for sustainable use and remediation of contaminated land.

Keywords: Bacterial communities - Heavy-metals - Hyperaccumulators - Nickel resistance - Serpentine soil

Received for publication March 25, 2009. In the natural environment, minerals are often associated with coexisting microorganisms. These interactions have profound impacts on the fate of a wide variety of contaminants. However, little information is available on the sorption of hydrophobic organic compounds (HOC), such as polycyclic aromatic hydrocarbons and chlorinated benzenes, onto the composites of minerals with bacteria, and knowledge of the influence of combined bacteria on HOC sorption to minerals is limited. In our study, sorption isotherms of phenanthrene (Phen) and 1,2,3-trichlorobenzene (TCB) onto *Bacillus subtilis*, minerals (kaolinite, montmorillonite, and goethite), and mineral–*B. subtilis* composites were studied to determine the role of *B. subtilis* in sorption. For pure mineral systems, the order of Phen and TCB sorption affinity was montmorillonite > kaolinite > goethite. For mineral–*B. subtilis* composites, the trend was montmorillonite > goethite > kaolinite, consistent with that of their ability to combine with bacteria. The coating of *B. subtilis* with minerals enhanced the sorption due to the strong sorption of Phen and TCB onto *B. subtilis* cells and the increase of total organic carbon of minerals. With increasing *B. subtilis* concentration, sorption of Phen and TCB on pure *B. subtilis* cells decreased, but sorption on kaolinite surface increased. Sodium azide can greatly reduce sorption capacity but increases sorption linearity for *B. subtilis* and mineral–*B. subtilis* composites. Compared with TCB, Phen had higher sorption affinity due to its high hydrophobicity. Our results may be useful for understanding the role of bacteria in regulating the distribution and transport of HOCs in the environment.

Abbreviations: HOCs, hydrophobic organic compounds • HPLC, high-performance liquid chromatography • PAHs, polycyclic aromatic hydrocarbons • Phen, phenanthrene • SEM, scanning electron microscopy • TCP, 1,2,3-trichlorobenzene • TOC, total organic carbon

Lei Yan[a], Huanhuan Yin[a], Shuang Zhang[b], Feifan Leng[a], Wenbin Nan[a] and Hongyu Li[a]. ([a] MOE Key Laboratory of Arid and Grassland Ecology, School of Life Sciences, Lanzhou University, 222 South Tianshui Road, Lanzhou 730000, PR China. [b] College of Life Science and Technology, Heilongjiang August First Land Reclamation University, Daqing 16331, PR China). **Effect of Combined *Bacillus subtilis* on the Sorption of Phenanthrene and 1,2,3-Trichlorobenzene onto Mineral Surfaces. Journal of Hazardous Materials, Volume 178 (1-3) (2010): 209-217**

The traditional techniques for removing low concentration arsenic are unsuitable. The biosorption characteristics of arsenite (iAs^{III}) and monomethyl arsonate (MMA^{V}) from aqueous

solution by *Acidithiobacillus ferrooxidans* BY-3 (*At. f* BY-3) were investigated as a function of pH, contact time, initial arsenic concentration, biomass dosage and temperature in this study. Results indicated that Langmuir isotherm model fitted better than Freundlich model to the equilibrium data. Analysis of kinetic data showed that the biosorption processes of both iAs^{III} and MMA^{V} involved pseudo-second-order kinetics. The thermodynamic parameters such as ΔG^{o}, ΔH^{o} and ΔS^{o} of the biosorption process showed that the adsorption of iAs^{III} and MMA^{V} onto *At. f* BY-3 was feasible, spontaneous and endothermic under the examined conditions. The competitive biosorption of iAs^{III} and MMA^{V} in binary mixture system was evaluated, and the results indicated that *At. f* BY-3 favored MMA^{V} biosorption. Fourier-transform infrared spectroscopy (FT-IR) showed –OH and –NH groups were involved in the biosorption process.

Keywords: Biosorption; Arsenite (iAs^{III}); Monomethyl arsonate (MMA^{V}); *Acidithiobacillus ferrooxidans*; Thermodynamics

Hao Chen [a], Guoliang Dai[a], Jie Zhao[a], Aiguo Zhong[a], Junyong Wu[a] and Hua Yan[a]. ([a] School of Pharmaceutical and Chemical Engineering, Taizhou University, Dongfang Road No. 605, Linhai 317000, Zhejiang, PR China). **Removal of copper(II) ions by a biosorbent—*Cinnamomum camphora* leaves powder. Journal of Hazardous Materials, Volume 177 (1-3) (2010): 228-236**

In the present study, *Cinnamomum camphora* leaves powder (CLP) was investigated as a biosorbent for the removal of copper ions from aqueous solutions. The biosorbents before and after adsorption were measured by EDS and FT-IR. Kinetic data and sorption equilibrium isotherms were carried out in batch process. The adsorption kinetic experiments revealed that there are three stages in the whole adsorption process. It was found that Cu(II) adsorption onto CLP for different initial Cu(II) concentrations all followed pseudo-second order kinetics and were mainly controlled by the film diffusion mechanism. Batch equilibrium results at different temperatures suggest that Cu(II) adsorption onto CLP can be described perfectly with Langmuir isotherm model compared to Freundlich and D–R isotherm models, and the characteristic parameters for each adsorption isotherm were also determined. Thermodynamic parameters calculated show that the adsorption process has been found to be endothermic in nature. The analysis for the values of the mean free energies of adsorption (E_a), the Gibbs free energy (ΔG^0) and the effect of ionic strength all demonstrate that the whole adsorption process is mainly dominated by ion-exchange mechanism, accompanied by a certain amount of surface complexation which has been verified by variations in EDS and FT-IR spectra and pH value before and after adsorption. Regeneration studies show CLP possesses an excellent reusability.

Keywords: Adsorption; *Cinnamomum camphora* leaf; Cu(II); Kinetics; Isotherms; Thermodynamics

D.K. Gupta[a], H.G. Huang[a], X.E. Yang[a], B.H.N. Razafindrabe[c] and M. Inouhe[b]. ([a] Ministry of Education Key Laboratory of Environmental Remediation and Ecosystem Health, Zhejiang University, Hua-jian-chi Campus, Hangzhou 310029, China. [b] Department of Biology and Environmental Sciences, Graduate School of Science and Engineering, Ehime University, Matsuyama, Ehime 790-8577, Japan. [c] Graduate School of Environment and Information Sciences, Yokohama National University, 79-7 Tokiwadai, Hodogaya Ku, 247-8501 Yokohama, Japan). **The detoxification of lead in *Sedum alfredii* H. is not related to phytochelatins but the glutathione. Journal of Hazardous Materials, Volume 177 (1-3) (2010): 437-444**

Two ecotypes of *S. alfredii* [Pb accumulating (AE) and Pb non-accumulating (NAE)] differing in their ability in accumulating Pb were exposed to different Pb levels to evaluate the effects on plant length, photosynthetic pigments, antioxidant enzymes (SOD and APX), cysteine, non-protein thiols (NP-SH), phytochelatins (PCs) and glutathione (GSH) vis-à-vis Pb accumulation. Both ecotypes showed significant Pb accumulation in roots, however only the AE showed significant Pb accumulation in shoots. We found that both AE and NAE of *S. alfredii*-induced biosynthesis of GSH rather than phytochelatins in their tissue upon addition of even high Pb levels (200 µM). Root and shoot length were mostly affected in both ecotypes after addition of higher Pb concentrations and on longer durations, however photosynthetic pigments did not alter upon addition of any Pb treatment. Both superoxide dismutase (SOD) and ascorbate peroxidase (APX) activities of AE were higher than NAE. The levels of cysteine and NP-SH were also higher in AE than in NAE. Hence, the characteristic Pb accumulation of ecotypes differed presumably in relation to their capacity for detoxification of Pb. These results suggest that enzymatic and non-enzymatic antioxidants play a key role in the detoxification of Pb-induced toxic effects in *Sedum alfredii*. This plant can be used as an indicator species for Pb contamination.

Keywords: Ascorbate peroxidase; Glutathione; Lead; Phytochelatins; Superoxide dismutase; *Sedum alfredii*

Carolina Prado[a, 1], Luisa Rodríguez-Montelongo[b], Juan A. González[c], Eduardo A. Pagano[d], Mirna Hilal[a] and Fernando E. Prado[a], . ([a] Cátedra de Fisiología Vegetal, Facultad de Ciencias Naturales e IML, Universidad Nacional de Tucumán, CP 4000, Tucumán, Argentina. [b] Departamento Bioquímica de la Nutrición, Instituto Superior de Investigaciones Biológicas (CONICET-UNT), CP 4000, Tucumán, Argentina. [c] Instituto de Ecología, Fundación Miguel Lillo, CP 4000, Tucumán, Argentina. [d] Departamento de Biología Aplicada y Alimentos, Facultad de Agronomía, Universidad de Buenos Aires, CP 1417, Buenos Aires, Argentina). **Uptake of chromium by *Salvinia minima*: Effect on plant growth, leaf respiration and carbohydrate metabolism . Journal of Hazardous Materials, Volume 177 (1-3) (2010): 546-553**

Metabolic responses to chromium (Cr) exposure and metal uptake were investigated using *Salvinia minima* plants. Cr treatment reduced the dry weight of floating and submerged leaves, while photosynthetic pigments were not affected. Measurements of respiratory oxygen uptake with and without inhibitors (KCN and SHAM) demonstrated that total respiration, alternative oxidase capacity and residual respiration were higher in Cr-treated than in Cr-untreated leaves, but the highest values were observed in floating leaves. Cr affected the soluble sugar content. Sucrose concentration was, in general, higher in Cr-treated than in Cr-untreated leaves, while the glucose concentration showed an inverse pattern. Cr also affected soluble acid invertase activity, but affectation trend was different between both leaves. Highest values of invertase activity were observed in Cr-treated floating leaves. According to our data soluble acid invertase and sucrose seem to be related to alternative oxidase capacity and residual respiration in floating and submerged leaves exposed to Cr. Thereby, this study constitutes an important contribution to understand metabolic relationships between mitochondrial respiration, alternative respiratory pathway and soluble carbohydrates in plants exposed to heavy metals.

Keywords: Alternative respiration; Carbohydrates; Cr; Invertase; *Salvinia minima*

Qingqing Peng[a, b], Yunguo Liu[a, b], Guangming Zeng[a, b], Weihua Xu[a, b], Chunping Yang[a, b] and Jingjin Zhang[a, b]. ([a] College of Environmental Science and Engineering, Hunan University, Changsha 410082, PR China. [b] Key Laboratory of Environmental Biology and Pollution Control (Hunan University), Ministry of Education, Changsha 410082, PR China). **Biosorption of copper(II) by immobilizing *Saccharomyces cerevisiae* on the surface of chitosan-coated magnetic nanoparticles from aqueous solution. Journal of Hazardous Materials, Volume 177 (1-3) (2010): 676-682**

Immobilized *Saccharomyces cerevisiae* on the surface of chitosan-coated magnetic nanoparticles (SICCM) was applied as a new magnetic adsorbent for the adsorption of Cu(II) from aqueous solution. The prepared magnetic adsorbent was characterized by TEM, XRD and FTIR. TEM images indicated that *S. cerevisiae* was immobilized on the surface of chitosan-coated magnetic nanoparticles (CCM) successfully, and conglobation was not observed. The XRD pictures suggested that the Fe_3O_4 nanoparticles were pure Fe_3O_4 with a spinel structure and that the immobilizing process did not result in the phase change of Fe_3O_4. Factors that influence the adsorption of Cu(II) were investigated, which included the initial pH of Cu(II) solution, initial concentration of Cu(II) solution and contact time. The optimum pH for Cu(II) absorption was 4.5. The highest removal efficiency of 96.8% was reached when the initial Cu(II) concentration was 60 mg L^{-1}, and the adsorption capacity was increased with the increase of initial concentration of Cu(II). In particular, SICCM was highly efficient for the fast adsorption of Cu(II) within the first 10 min, and adsorption equilibrium could be achieved in 1 h. Equilibrium studies show that the data of Cu(II) adsorption follow the Langmuir model. The maximum adsorption capacity for Cu(II) was estimated to be 144.9 mg g^{-1} with a Langmuir adsorption equilibrium constant of 0.0719 L mg^{-1} at 301 K.

Keywords: *Saccharomyces cerevisiae*; Chitosan-coated magnetic nanoparticle; Immobilization; Cu(II); Adsorption

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Organic pollutants in sediments are a worldwide problem because sediments act as sinks for hydrophobic, recalcitrant and hazardous compounds. Depending on biogeochemical processes these hydrocarbons are involved in adsorption, desorption and transformation processes and can be made available to benthic organisms as well as organisms in the water column through the sediment–water interface. Most of these recalcitrant hydrocarbons are toxic and carcinogenic, they may enter the food-chain and accumulate in biological tissue. Several approaches are being investigated or have been already used to remove organic hydrocarbons from sediments. This paper provides a review on types and sources of organic pollutants as well as their behavior in sediments. It presents the advantages and disadvantages of traditional sediment remediation techniques in use, such as dredging, capping and monitored natural attenuation. Furthermore, it describes new approaches with emphasis on bioremediation, like biostimulation, bioaugmentation and phytoremediation applied to sediments. These new techniques promise to be of lower impact and more cost efficient than traditional management strategies.

Keywords: Sediment; Organic pollutants; Remediation technologies; Bioremediation

C.-H. Chen[a], C.-F. Chang[a] and S.-M. Liu[a]. ([a] Institute of Marine Biology, National Taiwan Ocean University, 2-Peining Rd., Keelung, Taiwan). Partial degradation mechanisms of malachite green and methyl violet B by *Shewanella decolorationis* NTOU1 under anaerobic conditions. Journal of Hazardous Materials, Volume 177 (1-3) (2010): 281-289

This work demonstrated that *Shewanella decolorationis* NTOU1 decolorized 200 mg l^{-1} of crystal violet, malachite green, or methyl violet B within 2–11 h under anaerobic conditions at 35 °C. The initial color removal rate of malachite green was highest, while that of methyl violet was lowest. GC/MS analyses of the intermediate compounds produced during and after decolorization of malachite green and methyl violet B suggested that biodegradation of these dyes involved reduction to leuco form, N-demethylation, and reductive splitting of the triphenyl rings. The number of N-methylated groups of these dyes might have influenced decolorization rates and the reductive splitting of the triphenyl rings of these dyes. Cytotoxicity and antimicrobial test data showed that malachite green and methyl violet B solution (100 mg l^{-1}) were toxic. Toxicity of the dyes decreased after their decolorization, but further incubation resulted in increased toxicity.

Keywords: Decolorization; Malachite green; Methyl violet B; Partial degradation mechanisms; *Shewanella decolorationis* NTOU1

S.H. Hasan[a], P. Srivastava[a] and M. Talat[b]. ([a] Water Pollution Research Laboratory, Department of Applied Chemistry, Institute of Technology, Banaras Hindu University, Varanasi 221005, India. [b] Department of Biochemistry, Faculty of Science, Banaras Hindu University, Varanasi 221005, India). Biosorption of lead using immobilized *Aeromonas hydrophila* biomass in up flow column system: Factorial design for process optimization. Journal of Hazardous Materials Volume, 177 (1-3) (2010): 312-322

Free and immobilized biomass of *Aeromonas hydrophila* has been utilized for the removal of Pb(II) from aqueous solution. Fitness of Langmuir sorption model to the sorption data indicated the sorption was monolayer and uptake capacity of biomass was 163.9 and 138.88 mg/g for the free and immobilized biomass respectively. 85.38% Pb(II) removal was achieved at bed height of 19 cm and flow rate of 2 mL/min and BDST model was in a good agreement with the experimental results ($r^2 > 0.997$). An attempt has been made to optimize the process conditions for the maximum removal using Central Composite Design with the help of Minitab® 15 software and the result predicted by optimization plots was 88.27% which is close to the experimental data i.e. 85.38%. Sorption–desorption studies revealed that polysulfone immobilized biomass could reused up to 16 cycles and bed was completely exhaust after 33 cycles.

Keywords: *Aeromonas hydrophila*; Pb(II); Bed Depth Service Time (BDST) Model; Central Composite Design (CCD); Sorption–desorption

M. Dary[a], M.A. Chamber-Pérez[b], A.J. Palomares[a, b, 1] and E. Pajuelo[a]. ([a] Departamento de Microbiología y Parasitología, Facultad de Farmacia, Universidad de Sevilla. c/Profesor García González, 2, 41012 Seville, Spain. [b] Centro Las Torres, IFAPA (Junta de Andalucía), 41200 Seville, Spain). "*In situ*" phytostabilisation of heavy metal polluted soils

using *Lupinus luteus* inoculated with metal resistant plant-growth promoting rhizobacteria. Journal of Hazardous Materials, Volume 177 (1-3) (2010): 323-330

The aim of this work is the evaluation of metal phytostabilisation potential of *Lupinus luteus* inoculated with *Bradyrhizobium* sp. 750 and heavy metal resistant PGPRs (plant-growth promoting rhizobacteria), for *in situ* reclamation of multi-metal contaminated soil after a mine spill. Yellow lupines accumulated heavy metals mainly in roots (Cu, Cd and especially Pb were poorly translocated to shoots). This indicates a potential use of this plant in metal phytostabilisation. Furthermore, As accumulation was undetectable. On the other hand, zinc accumulation was 10–100 times higher than all other metals, both in roots and in shoots. Inoculation with *Bradyrhizobium* sp. 750 increased both biomass and nitrogen content, indicating that nitrogen fixation was effective in soils with moderate levels of contamination. Co-inoculation of lupines with a *consortium* of metal resistant PGPR (including *Bradyrhizobium* sp., *Pseudomonas* sp. and *Ochrobactrum cytisi*) produced an additional improvement of plant biomass. At the same time, a decrease in metal accumulation was observed, both in shoots and roots, which could be due to a protective effect exerted on plant rhizosphere. Our results indicate the usefulness of *L. luteus* inoculated with a bacterial consortium of metal resistant PGPRs as a method for *in situ* reclamation of metal polluted soils.

Keywords: Mine spill; *In situ* phytostabilisation; Phytoextraction; *Legume*; *Bradyrhizobium*; PGPR

Amit Bafana [a], Kannan Krishnamurthi[a], Mahendra Patil[a] and Tapan Chakrabarti[a]. ([a] Environmental Biotechnology Division, National Environmental Engineering Research Institute, Nehru Marg, Nagpur 440020, India). Heavy metal resistance in *Arthrobacter ramosus* strain G2 isolated from mercuric salt-contaminated soil. Journal of Hazardous Materials, Volume 177 (1-3) (2010): 481-486

Present study describes isolation of a multiple metal-resistant *Arthrobacter ramosus* strain from mercuric salt-contaminated soil. The isolate was found to resist and bioaccumulate several metals, such as cadmium, cobalt, zinc, chromium and mercury. Maximum tolerated concentrations for above metals were found to be 37, 525, 348, 1530 and 369 μM, respectively. The isolate could also reduce and detoxify redox-active metals like chromium and mercury, indicating that it has great potential in bioremediation of heavy metal-contaminated sites. Chromate reductase and mercuric reductase (MerA) activities in protein extract of the culture were found to be 2.3 and 0.17 units mg^{-1} protein, respectively. MerA enzyme was isolated from the culture by $(NH_4)_2SO_4$ precipitation followed by dye affinity chromatography and its identity was confirmed by nano-LC–MS/MS. Its monomeric molecular weight, and optimum pH and temperature were 57 kDa, 7.4 and 55 °C, respectively. Thus, the enzyme was mildly thermophilic as compared to other MerA enzymes. K_m and V_{max} of the enzyme were 16.9 μM $HgCl_2$ and 6.2 μmol min^{-1} mg^{-1} enzyme, respectively. The enzyme was found to be NADPH-specific. To our knowledge this is the first report on characterization of MerA enzyme from an *Arthrobacter* sp.

Keywords: Bioaccumulation; Chromate reductase; Mercuric reductase; MerA; Nano-LC–MS/MS

G. Mohanakrishna[a], S. Venkata Mohan [a] and P.N. Sarma[a]. ([a] Bioengineering and Environmental Centre (BEEC), Indian Institute of Chemical Technology (IICT),

Hyderabad 500 607, India). Bio-electrochemical treatment of distillery wastewater in microbial fuel cell facilitating decolorization and desalination along with power generation. Journal of Hazardous Materials, Volume 177 (1-3) (2010): 487-494

Microbial fuel cell (MFC; open-air cathode) was evaluated as bio-electrochemical treatment system for distillery wastewater during bioelectricity generation. MFC was operated at three substrate loading conditions in fed-batch mode under acidophilic (pH 6) condition using anaerobic consortia as anodic-biocatalyst. Current visualized marked improvement with increase in substrate load without any process inhibition (2.12–2.48 mA). Apart from electricity generation, MFC documented efficient treatment of distillery wastewater and illustrated its function as an integrated wastewater treatment system by simultaneously removing multiple pollutants. Fuel cell operation yielded enhanced substrate degradation (COD, 72.84%) compared to the fermentation process (~29.5% improvement). Interestingly due to treatment in MFC, considerable reduction in color (31.67%) of distillery wastewater was also observed as against color intensification normally observed due to re-polymerization in corresponding anaerobic process. Good reduction in total dissolved solids (TDS, 23.96%) was also noticed due to fuel cell operation, which is generally not amenable in biological treatment. The simultaneous removal of multiple pollutants observed in distillery wastewater might be attributed to the biologically catalyzed electrochemical reactions occurring in the anodic chamber of MFC mediated by anaerobic substrate metabolism.

Keywords: Open-air cathode; Anaerobic mixed consortia; Electrochemical oxidation; Electrolytic dissociation; TDS; Color

Abbreviations: AB, acidogenic bacteria; C, charge (C); C_D, current density (mA/m^2); COD, chemical oxygen demand (mg/l); C_S, COD/carbohydrate/turbidity concentration in outlet of MFC; C_{SO}, COD/carbohydrate/turbidity concentration in feed to MFC; CV, cyclic voltammetry; DAO, direct anodic oxidation; e^-, electron; emf, electro motive force (V); ETP, effluent treatment plant; $E_{o,anodic}$, anodic potential; H^+, proton; I, current (mA); IO, indirect oxidation; MA, methanogenic archaea; MFC, microbial fuel cell; OCV, open circuit voltage (V); OL, organic/substrate loading (kg COD/m^3); OM, organic matter/substrate; P, power (mW); P_D, power density (mW/m^2); PEM, proton exchange membrane; RDAP, relative decrease in anodic potential (%); RE, reference electrode (Ag/AgCl); SD, substrate degradation (kg COD_R/m^3); SPY, specific power yield (W/kg COD_R); TDS, total dissolved solids (mg/l); V, voltage (V); VFA, volatile fatty acids (mg/l); VPP, volumetric power production (mW/m^3); VSS, volatile suspended solids (mg/l); ξ_{COD}, COD removal efficiency (%); $\xi_{Turbidity}$, turbidity removal efficiency (%); $\xi_{Carbohydrate}$, carbohydrate removal efficiency (%); ξ_{Color}, color removal efficiency (%)

A.R. Binupriya[a], M. Sathishkumar[b], K. Vijayaraghavan[b] and S.-I. Yun[a]. ([a] Department of Food Science and Technology, College of Agriculture and Life Science, Chonbuk National University, Jeonju 561-756, Republic of Korea. [b] Singapore-Delft Water Alliance, National University of Singapore, 2 Engineering Drive 2, Singapore 117577, Singapore). **Bioreduction of trivalent aurum to nano-crystalline gold particles by active and inactive cells and cell-free extract of *Aspergillus oryzae* var. *viridis*. Journal of Hazardous Materials, Volume 177 (1-3) (2010): 539-545**

Bioreduction efficacy of both active (AB) and inactive (IB) cells/biomass of *Aspergillus oryzae* var. *viridis* and their respective cell-free extracts (ACE and ICE) to convert trivalent aurum to gold nanoparticles were tested in the present study. Strong plasmon resonance of gold nanoparticles was observed between 540 and 560 nm in the samples obtained from AB, IB, ACE and ICE. Transmission electron microscopy (TEM), field emission scanning electron microscopy (FE-SEM), energy dispersive X-ray (EDX) and X-ray diffraction (XRD) were performed to examine the formation of gold nanoparticles. Comparing all four forms of *A. oryzae* var. *viridis*, ICE showed high gold nanoparticle productivity. The nanoparticles formed were quite uniform in shape and ranged in size from 10 to 60 nm. In addition some triangle, pentagon and hexagon-shaped nanoplates with size range of 30–400 nm were also synthesized especially at lower pH. Organics from the inactive cells are believed to be responsible for reduction of trivalent aurum to nano-sized gold particles. Organic content of the ICE was found to be double the amount of ACE. High productivity of gold nanoparticles by metabolic-independent process opens up an interesting area of nanoparticle synthesis using waste fungal biomass from industries.

Keywords: *Aspergillus oryzae* var. *viridis*; Active and inactive biomass; Cell-free extract; Bioreduction; Gold nanoparticles

Abbreviations: AB, active biomass; IB, inactive biomass; ACE, active cell extract; ICE, inactive cell extract; Au NPs, gold nanoparticles

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The present work is concerned with the defluoridation capacities of activated (ATFS) and MnO_2-coated Tamarind Fruit Shell (MTFS), using batch and column sorption techniques. In the batch technique, the dynamics of fluoride sorption, with respect to pH, $[F]_o$ and sorbent dose, was studied. The applicability of pseudo-first order for ATFS and Ritchie-second order for MTFS was observed. The kinetics data were found to fit well with Temkin isotherm for ATFS and Langmuir for MTFS. The interaction of co-ions in the defluoridation capacity of the sorbent was studied. Column experiments were carried out under a constant fluoride concentration of 2 mg/l, flow rate and different bed depths. The capacities of the breakthrough and exhaustion points increased with increase in the bed depth for ATFS unlike MTFS. The Thomson model was applied to the column experimental results. The characterization of the sorbents, ATFS and MTFS, was done using the FTIR, SEM and XRD techniques.

Keywords: Defluoridation; Activated Tamarind Fruit Shell; Manganese oxide-coated Tamarind Fruit Shell; FTIR; SEM; XRD

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Laboratory of Microbiological Engineering of Agricultural Environment, Ministry of Agriculture, Nanjing Agricultural University, Nanjing, 210095, Jiangsu, People's Republic of China). Bioaugmentation of a sequencing batch reactor with *Pseudomonas putida* ONBA-17, and its impact on reactor bacterial communities. Journal of Hazardous Materials, Volume 176 (1-3) (2010): 20-26

This study demonstrates the feasibility of using *Pseudomonas putida* ONBA-17 to bioaugment a sequencing batch reactor (SBR) treating *o*-nitrobenzaldehyde (ONBA) synthetic wastewater. To monitor its survival, the strain was chromosomally marked with *gfp* gene. After a transient adaptation, almost 100% degradation of ONBA was obtained within 8 days as compared with 23.47% of the non-inoculated control. The bioaugmented reactor has a better chemical oxygen demand (COD) removal performance (96.28%) than that (79.26%) of the control. The bioaugmentation not only enhanced the removal capability of target compound, but shortened system start-up time. After the increase in ONBA load, performance fluctuation of two reactors was observed, and the final treating effects of them were comparable. What is more, denaturing gradient gel electrophoresis (DGGE) analysis of 16S rRNA genes via a combination of pattern comparison and sequence phylogenetic analysis was performed to uncover changes in sludge microbial communities. Only the members of alpha, beta and gamma subdivisions of Proteobacteria were identified. To isolate ONBA-degrading relevant microorganisms, spread plate was used and four bacterial strains were obtained. Subsequent systematic studies on these bacteria characterized their traits which to some extent explained why such bacteria could be kept in the system. This study will help future research in better understanding of the bioreactor bioaugmentation.

Keywords: Bioaugmentation; *o*-Nitrobenzaldehyde; *Pseudomonas putida*; DGGE; Loading shock

Ta-Chen Lin[a], Po-Tsen Pan[b] and Sheng-Shung Cheng[b, c]. ([a] Department of Biological Science and Technology, Meiho Institute of Technology, Pingtung County, 912, Taiwan. [b] Department of Environmental Engineering, National Cheng Kung University, Tainan City 701, Taiwan. [c] Sustainable Environment Research Center, National Cheng Kung University, Tainan City 701, Taiwan). Ex situ bioremediation of oil-contaminated soil. Journal of Hazardous Materials, Volume 176 (1-3) (2010): 27-34

An innovative bioprocess method, Systematic Environmental Molecular Bioremediation Technology (SEMBT) that combines bioaugmentation and biostimulation with a molecular monitoring microarray biochip, was developed as an integrated bioremediation technology to treat S- and T-series biopiles by using the landfarming operation and reseeding process to enhance the bioremediation efficiency. After 28 days of the bioremediation process, diesel oil ($TPH_{C10–C28}$) and fuel oil ($TPH_{C10–C40}$) were degraded up to approximately 70% and 63% respectively in the S-series biopiles. When the bioaugmentation and biostimulation were applied in the beginning of bioremediation, the microbial concentration increased from approximately 10^5 to 10^6 CFU/g dry soil along with the TPH biodegradation. Analysis of microbial diversity in the contaminated soils by microarray biochips revealed that *Acinetobacter* sp. and *Pseudomonas aeruginosa* were the predominant groups in indigenous consortia, while the augmented consortia were *Gordonia alkanivorans* and *Rhodococcus erythropolis* in both series of biopiles during bioremediation. Microbial respiration as influenced by the microbial activity reflected directly

the active microbial population and indirectly the biodegradation of TPH. Field experimental results showed that the residual TPH concentration in the complex biopile was reduced to less than 500 mg TPH/kg dry soil. The above results demonstrated that the SEMBT technology is a feasible alternative to bioremediate the oil-contaminated soil.

Keywords: Hydrocarbon; Landfarming; Bioremediation; Bioaugmentation; Biostimulation

Amparo Mauricio Gutiérrez[a]; Juan José Peña Cabriales[a]; María Maldonado Vega[b]. ([a] Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional Campus Guanajuato, Irapuato, Guanajuato, México. [b] Dirección en Investigación Ambiental, Centro de Innovación Aplicada en Tecnologías Competitivas A. C., León, Guanajuato, México)**Isolation and Characterization of Hexavalent Chromium-Reducing Rhizospheric Bacteria From a Wetland. International Journal of Phytoremediation, Volume 12 (4) (2010): 317 – 334**

Scirpus americanus Pers. occurs naturally in "San Germán," a pond that serves as a receptor of industrial wastewater in Guanajuato, México. This plant accumulates metals mainly in the root: concentrations (mg/kg) of Cr, As, Cd and Se were 970, 49, 41, and 85 respectively. Analysis of rhizosphere samples indicated bacterial population of 10^8 cfu g^{-1} in media with 0.2 mM Cr(VI) and 10 mM sodium gluconate. Thirteen isolates were obtained and phylogenetic analyses (16S rRNA) indicated they corresponded to genera of *Agrobacterium, Arthrobacter*, *Microbacterium*, *Curtobacterium, Rhodococcus, Xanthomonas* and *Pseudomonas*. Cr(VI) reduction was evaluated using the diphenyl carbazide method. The isolates accomplished 5-40% (20 µM) of reduction in assays of resting cell and tolerated 0.5-5.0 mM Cr(VI). Eight strains used nitrate and thirteen used iron and chromium as electron acceptors to grow under anaerobic conditions. Cr(VI) reduction by five strains occurred at pH values (7-9) and NaCl concentrations (0.5-1.0 M) in basal medium. A mixed culture of strains (S17 and S28) reached a chromium removal of 100% at 0.2 mM Cr(VI) initial concentration. Aerobically, this consortium was capable of 93.8% Cr(VI) reduction of 81 μgL^{-1} Cr(VI) of the industrial effluent, indicating their possible use in environmental cleanup.

Keywords: Bioremediation; bacterial consortia; Scirpus americanus Pers

Wuxing Liu[a]; Yongming Luo[a]; Ying Teng[a]; Zhen'gao Li[a]. ([a] Key Laboratory of Soil Environment and Pollution Remediation, Institute of Soil Science, Chinese Academy of Sciences, Nanjing, P.R. China). **Phytoremediation of Oilfield Sludge After Prepared Bed Bioremediation Treatment. International Journal of Phytoremediation, Volume 12 (3) (2010): 268 – 278**

A field-scale experiment was carried out to compare the effectiveness of five plant species in the remediation of oily sludge. Alfalfa, tall fescue, and soybean substantially increased the removal rate of oil and grease (O&G) after 120 days of remediation. Of these, soybean treatment showed the highest removal rate of 34.2% compared with only 13.7% in the unplanted control plot. In addition to assisting with phytoremediation, soybean can also be used for energy production, for example in the production of biodiesel. Furthermore, microbial counts and community level physiological profiling using Biolog sole carbon source utilization tests were used to investigate the effect of plants on the microbial community of oily sludge. Plants selectively increased microbial counts in the rhizosphere and O&G concentrations of the sludge were negatively correlated with counts of TPH (total petroleum hydrocarbon) degraders. The acute biotoxicity of

the sludge was also tested by the bacterial luminescence method and alfalfa treatments decreased the biological toxicity of the sludge compared with the unplanted control.

Keywords: oily sludge; phytoremediation; microbial diversity; biodiesel

Jan Kotyza[ab]; Petr Soudek[a]; Zdeněk Kafka[b]; Tomáš Vaněk[a]. ([a] Laboratory of Plant Biotechnologies, Joint Laboratory of Institute of Experimental Botany and Institute of Crop Research, Prague, Czech Republic. [b] Faculty of Environment Technology, Institute of Chemical Technology, Prague, Czech Republic). Phytoremediation of Pharmaceuticals—Preliminary Study. International Journal of Phytoremediation, Volume 12 (3) (2010): 306 – 316

Phytoremediation of selected pharmaceuticals (diclofenac, ibuprofen, and acetaminophen) using *Armoracia rusticana* and Linum **usitatissimum** cell cultures and by hydroponically cultivated *Lupinus albus*, *Hordeum vulgaris*, and *Phragmites australis* plants in laboratory conditions is described. During in vitro experiments, the best results for acetaminophen were achieved using Armoracia rusticana hairy root cultures, where 100% of the starting amount was removed from the media during eight days. Total removal of ibuprofen and diclofenac was achieved using a *Linum usitatissimum* suspension culture after one and six days, respectively. In the hydroponic arrangement, the best results were achieved for *Lupinus*, where acetaminophen was totally removed from media during two or four days in concentrations of 0.1 or 0.2 mM, respectively. The best effectiveness of ibuprofen removal (50% of starting amount) was found in case of *Phragmites*. Effectiveness of all tested plants for diclofenac removal was low. The best removal was achieved using Phragmites in the case of 0.2 mM concentration—67% of the starting amount and Hordeum for 0.1 mM starting concentration, 56%.

Keywords: acetaminophen; diclofenac; ibuprofen; paracetamol; pharmaceuticals; phytoremediation; wastewater

A. A. Romeh[a]. ([a] Plant Production Department, Efficient Productivity Institute, Zagazig University Zagazig, Egypt). Phytoremediation of Water and Soil Contaminated with Imidacloprid Pesticide by *Plantago Major*, L. International Journal of Phytoremediation, Volume 12 (2) (2010): 188 - 199

Broadleaf plantain plant (Plantago major L.) was used in phytoremediation of imidacloprid insecticide in water and soils. For the Freundlich model, the constant related to the biosorption capacity (K_f) of imidaclaprid were respectively, 7.94, 6.31, and 2.51ug/g for dry roots, fruits (seeds with shells) and leaves of broadleaf plantain plant. Viable whole broadleaf plantain plant in water solution reduced imidacloprid residues by 55.81-95.17%, during 1-10 days of exposure periods compared with 13.71-61.95% in water solution without the plantain. In water solution, imidacloprid significantly accumulated in plantain roots, leaves and fruits to reach the maximum levels after 6, 1 and 3 days of treatment, respectively. The maximum levels were 15.74, 37.21, and 5.74 ug/gm, respectively. These values were decreased to 6.95, 1.46, and 0.12 ug/ gm after 10 days of treatment. The growing cells of short-rod gram-negative bacteria that isolated from the water solution containing broadleaf plantain plants was able to induce 93.34% loss of imidacloprid as a source of both carbon and nitrogen within a short period (48 hr) compared with

31.90% in un inoculated medium. Half-life (t 1/2) in soil planted with broadleaf plantain plants and in unplanted soil were found to be 4.8 and 8.4 days, respectively.

Keywords: Pytoremediation; broadleaf plantain plant; soil; water; imidacloprid

Jennifer Read[a]; Tim D. Fletcher[b]; Tricia Wevill[a]; Ana Deletic[b]. ([a] Facility for Advancing Water Biofiltration-FAWB, School of Biological Sciences, Monash University, Victoria, Australia. [b] Facility for Advancing Water Biofiltration-FAWB, Department of Civil Engineering, Monash University, Victoria, Australia). **Plant Traits that Enhance Pollutant Removal from Stormwater in Biofiltration Systems. International Journal of Phytoremediation, Volume 12 (1) (2010): 34 – 53**

Plants species have been shown to improve the performance of stormwater biofiltration systems, particularly in removal of N and P. Recent research has shown that plants vary in their contribution to pollutant removal, but little is known about the type of plant that is best suited to use in biofilters in terms of survival, growth rate, and performance. In this study, growth responses of 20 species to applications of semi-synthetic stormwater were measured, and the roles of key plant traits in removal of N, P, and several metals were investigated. There was no evidence of negative effects of stormwater application on plant growth, and plant traits, particularly root traits, were strongly correlated negatively with N and P concentrations of effluent stormwater. The most common and strong contributors to N and P removal appeared to be the length of the longest root, rooting depth, total root length, and root mass. The plants that made the strongest contribution to pollutant removal, *e.g.*, *Carex appressa*, combined these traits with high growth rates. Investigation of other plant traits (*e.g.*, physiology), causal mechanisms, and effects of more complex planting environments (*e.g.*, *species mixtures)* should further guide the selection of plants to enhance performance of biofiltration systems.

Keywords: Carex; metals; nitrogen; phosphorus; roots; runoff

Liu Xingyu[a, b], Wu Biao[a], Chen Bowei[a], Wen Jiankang[a], Ruan Renman[b], Yao Guocheng[a] and Wang Dianzuo[c]. ([a] National Engineering Laboratory of Biohydrometallurgy, General Research Institute for Nonferrous Metals, No.2 Xinjiekuowai Street, Beijing 100088, China. [b] Zijin Mining Group Co., Ltd, Fujian, China. [c] Chinese Academy of Engineering, Beijing, China). **Bioleaching of chalcocite started at different pH: Response of the microbial community to environmental stress and leaching kinetics. Hydrometallurgy, Volume 103 (1-4) (2010): 1-6**

Bioleaching of chalcocite mineral was conducted in two stirred reactors at 30 °C. Bacteria culture from the Zijin copper mine bio-heap was applied in this test. Leaching test started from pH 1.2 was carried in reactor A, and for reactor B, a start-up pH of 1.5 was used. Results showed that low pH could effectively facilitate copper dissolution and the affected microbial community structure thus inhibited iron dissolution. By using the 16 S rRNA gene clone library technique, bacterial community structure and change in the two reactors was revealed. In the inoculated culture, the dominant bacteria groups were *Leptospillium*, *Acidithiobacillus* and *Sulfobacillus*. While as leaching tests continued, the bacteria community in the two reactors presented different structures. A decrease in the proportion of *Leptospillium* from 22.2% to 10% and an increase in the proportion of *Acidithiobacillus* from 31.1% to 86% was detected in reactor A under relative higher redox potential (from 651 mV to 705 mV) and low pH (pH1.2). While for reactor B, an increase in the proportion of *Leptospillium* and *Acidithiobacillus* under relative low redox

potential (from 530 mV to 631 mV) and high pH (pH 1.5–1.6) was observed, each from 22.2% to 59.2% and 31.1% to 40.8% respectively. Leaching experiment data were fitted to the shrinking core model. Diffusion control was found to be more significant at pH 1.2 bioleaching than it was at pH 1.5 bioleaching. These results may help us understand bioleaching behavior of secondary copper sulfide and improve commercial plant operation.

Keywords: Bioleaching; Biodiversity; Chalcocite; 16 S rRNA gene

J. Mann[1], J. L. Markham[2], P. Peiris[2], N. Nair[1], R. N. Spooner-Hart[1] and P. Holford[1]. ([1]Centre for Plants and the Environment, University of Western Sydney, Locked Bag 1797, Penrith South DC, NSW, 1797, Australia. [2]School of Natural Sciences, University of Western Sydney, Penrith South DC, NSW, Australia). **Screening and selection of fungi for bioremediation of olive mill wastewater. World Journal of Microbiology and Biotechnology, Volume 26 (3) (2010): 567-571**

Olive mill wastewater (OMWW) is a significant pollutant and its phytotoxicity is attributed mostly to the phenols present. 220 fungi were screened for their ability to produce detoxifying enzymes and/or grow in OMWW. Four isolates, species of *Cerrena, Byssochlamys*, *Lasiodiplodia* and *Bionectria* were selected and compared against *Phanerochaete chrysosporium* for their ability to bioremediate OMWW in the presence of a competing indigenous microflora. For the first time we report that a *Cerrena* sp. achieved a 75% reduction of phenolics in OMWW and that, unusually, the reduction occurred within 2 h of the addition to the OMWW.

Keywords: Olive mill wastewater - Fungi - *Cerrena* sp. - Bioremediation - Phenols - COD - Phytotoxicity

Somayeh Niknam[1], Mohammad Ali Faramarzi[1, 2], Khosrou Abdi[3], Mojtaba Tabatabaei Yazdi[1], Mohsen Amini[3] and Hossein Rastegar[4]. ([1]Department of Pharmaceutical Biotechnology, Biotechnology Research Center, Faculty of Pharmacy, Tehran University of Medical Sciences, P.O. Box 14155-6451, 14174 Tehran, Iran. [2]Department of Medical Biotechnology, Faculty of Advanced Medical Technologies, Tehran University of Medical Sciences, 14174 Tehran, Iran. [3]Department of Medicinal Chemistry, Faculty of Pharmacy, Tehran University of Medical Sciences, 14174 Tehran, Iran. [4]Food & Drug Laboratory Research Center, Ministry of Health & Medical Education, Tehran, Iran). **Bioconversion of codeine to semi-synthetic opiate derivatives by the cyanobacterium *Nostoc muscorum*. World Journal of Microbiology and Biotechnology, Volume 26 (1) (2010): 119-123**

Very limited studies have been done to investigate the algal biotransformation of codeine to its opioid derivatives. On the other hand, microalgae have been recently introduced as potential tools for green synthesis of various organic compounds. In the present work, the capability of biotransformation of codeine by a locally isolate strain of cyanobacterium, *Nostoc muscorum*, was evaluated. Incubation of the whole cells of *Nostoc muscorum* with codeine (**I**) under continuous light photoregime of 60 µmol photons/m^{2}s at 25°C for 5 days gave rise to four transformation products. The bioproducts were separated by gas chromatography and identified as 6-acetylcodeine (**II**), oxycodone (**III**), norcodeine (**IV**), morphine (**V**) and based on their mass spectra. Observed modifications included O-demethylation, N-demethylation, C6-acetylation,

C14-hydroxilation, Δ^7-reduction, and C6-oxidation. The ability of *N. muscorum* to convert codeine to oxycodone (**III**) represents an uncommon pattern of codeine metabolism in microorganisms that may be of industrial importance.

Keywords: *Nostoc muscorum* - Biotransformation - Alkaloid - Codeine - Opiate derivatives

P.J. Strong[1] ([1]Scion, Private Bag 3020, Rotorua, 3046, New Zealand). Fungal remediation of Amarula distillery wastewater. World Journal of Microbiology and Biotechnology, Volume 26 (1) (2010): 133-144

Amarula Cream is an alcoholic beverage derived from the distillation of fermented marula fruit and to date there is no scientific data as to the characteristics of the distillery wastewater generated from its production. The wastewater was found to have a chemical oxygen demand (COD) of 27 g/l, a pH of 3.8, a high concentration of phenolic compounds (866 mg/l) and a high suspended solids content (10.5 g/l), all of which could adversely affect biological treatment. Full-strength wastewater was treated using shake-flask monocultures of four white rot fungi (*Trametes pubescens* MB 89, *Ceriporiopsis subvermispora*, *Pycnoporus cinnabarinus* or *Phanerochaete chrysosporium*) at pH 5.0 with no additional carbon or nitrogen supplements. *Trametes pubescens* performed the best with regards to degrading phenolic compounds, COD and colour, while *P. cinnabarinus* improved the pH to the greatest extent. Laccase synthesis was only detected in the *T. pubescens* and *C. subvermispora* cultures. Six wastewater concentrations (100, 80, 60, 40, 20 and 10%) were assessed at pH 4.5 to establish an optimum concentration for remediation and laccase production by *T. pubescens*. Similar COD removal efficiencies (71–77%) and phenolic removal efficiencies (87–92%) were achieved at all concentrations. The phenolic removal efficiencies improved by approximately 5% compared to the screening experiment at pH 5.0, indicating that the laccase was more efficient at pH 4.5. The pH became more basic as a result of treatment and the colour decreased for samples below 60% wastewater concentration. The maximum laccase activity (1063 ± 26 units/l) was obtained in the 80% wastewater concentration. This study has resulted in the first characterization of Amarula distillery wastewater and showed that it has a high phenolic compound concentration, COD and suspended solids content. It was possible to biologically treat the wastewater at full strength using a number of white-rot fungi just by raising the pH.

Keywords: Aerobic - Effluent - Enzyme - Laccase - Phenolic - Vinasse

Zandra Arwidsson[1, 2] and Bert Allard[1]. ([1]Man–Technology–Environment Research Centre, School of Science and Technology, Örebro University, 701 82 Örebro, Sweden. [2]SAKAB AB, 692 85 Kumla, Sweden). Remediation of Metal-Contaminated Soil by Organic Metabolites from Fungi II—Metal Redistribution. Water, Air, & Soil Pollution, Volume 207 (1-4) (2010): 5-18

Exudation of low molecular weight organic acids by fungi was studied in a project focusing on bioremediation of metal-contaminated soils. The production of acids (mainly oxalic and citric acid) as a response to nutrient variations and presence of metals has recently been reported (Arwidsson et al. *2009*). A significant release of metals was observed and was related not only to the production of organic acids but also to the resulting pH decrease in the systems. The processes governing the release and redistribution of metals in the soil–water fungus system were the focus of the present continuation of the project, based on observations of *Aspergillus niger*, *Penicillium bilaiae*, and a *Penicillium* sp. The release of lead was 12% from the soil with

the second highest initial load (1,600 mg kg^{-1}), while the release of copper was 90% from the same soil (140 mg kg^{-1}). The dominating mechanism behind the release and subsequent redistribution was the change in pH, going from near neutral to values in the range 2.1–5.9, reflecting the production of organic acids. For some of the systems, the formation of soluble complexes is indicated (copper, at intermediate pH) which favors the metal release. Iron is assumed to play a key role since the amount of secondary iron in the soils is higher than the total load of secondary heavy metals. It can be assumed that most of the heavy metals are initially associated with iron-rich phases through adsorption or coprecipitation. These phases can be dissolved, or associated metals can be desorbed, by a decrease in pH. It would be feasible to further develop a process in technical scale for remediation of metal-contaminated soil, based on microbial metabolite production leading to formation of soluble metal complexes, notably with copper.

Keywords: Bioremediation - Fungi - Metals - Oxalic acid - Citric acid

Johnson Kayode Adesodun[1], Mutiau O. Atayese[2], T. A. Agbaje[1], Bose A. Osadiaye[1], O. F. Mafe[1] and Adeniyi A. Soretire[1]. ([1]Department of Soil Science and Land Management, University of Agriculture, P.M. B 2240, Abeokuta, 110001, Ogun-State, Nigeria. [2]Department of Plant Physiology and Crop Production, University of Agriculture, P.M. B 2240, Abeokuta, 110001, Ogun-State, Nigeria). **Phytoremediation Potentials of Sunflowers (*Tithonia diversifolia* and *Helianthus annuus*) for Metals in Soils Contaminated with Zinc and Lead Nitrates. Water, Air, & Soil Pollution, Volume 207 (1-4) (2010): 195-201**

Two species of sunflower, i.e., *Tithonia diversifolia* and *Helianthus annuus*, were investigated for their potential to remove heavy metals from contaminated soils. Dried and mature *T. diversifolia* (Mexican flower) seeds were collected along roadsides, while *H. annuus* (sunflower) seeds were sourced from the Department of PBST, University of Agriculture Abeokuta, Nigeria. The contaminants were added as lead nitrate (Pb $(NO_3)_2$) and zinc nitrate (Zn $(NO_3)_2$) at 400 mg/kg which represents upper critical soil concentration for both Pb and Zn. The results indicated that *T. diversifolia* mopped up substantial concentrations of Pb in the above-ground biomass compared to concentrations in the roots. The concentrations in the leaf compartment were 87.3, 71.3, and 71.5 mg/kg at 4, 6, and 8 weeks after planting (AP), respectively. In roots, it was 99.4 mg/kg, 97.4 mg/g, and 77.7 mg/kg while 79.3, 77.8, and 60.7 mg/kg were observed in the stems at 4, 6, and 8 weeks AP, respectively. Observations with *H. annuus* followed the pattern found with *T. diversifolia,* showing significant ($p < 0.05$) accumulation of Pb in the above-ground biomass. Results obtained from Zn contaminated soils showed significant ($p < 0.05$) accumulation in the above-ground compartments of *T. diversifolia* and *H. annuus* compared with root. However, the highest accumulation of Zn was observed in the leaf. The translocation factor and enrichment coefficient of Pb and Zn with these plant species are greater than 1, indicating that these metals moved more easily in these plants. However, this result also showed that the translocation of Zn from root to the shoot of the two plants was higher than Pb. In conclusion, this experiment showed that these plants accumulated substantial Pb and Zn in their shoots (leaf and stem) at 4 weeks AP which diminished with time. This implies that the efficiency of these plants in cleaning the contaminated soils was at the early stage of their growth.

Keywords: Phytoremediation - Heavy metals - Sunflowers - Translocation factor - Enrichment coefficient

Zandra Arwidsson[1, 2], Emma Johansson[1], Thomas von Kronhelm[2], Bert Allard[1] and Patrick van Hees[1, 3]. ([1]Man-Technology-Environment Research Centre, School of Science and Technology, Örebro University, 701 82 Örebro, Sweden. [2]SAKAB AB, 692 85 Kumla, Sweden. [3]Eurofins Environment Sweden AB, Box 737, 531 17 Lidköping, Sweden). **Remediation of Metal Contaminated Soil by Organic Metabolites from Fungi I—Production of Organic Acids. Water, Air, & Soil Pollution, Volume 205 (1-4) (2010): 215-226**

Investigations were made on living strains of fungi in a bioremediation process of three metal (lead) contaminated soils. Three saprotrophic fungi (*Aspergillus niger*, *Penicillium bilaiae*, and a *Penicillium* sp.) were exposed to poor and rich nutrient conditions (no carbon availability or 0.11 M D-glucose, respectively) and metal stress (25 µM lead or contaminated soils) for 5 days. Exudation of low molecular weight organic acids was investigated as a response to the metal and nutrient conditions. Main organic acids identified were oxalic acid (*A. niger*) and citric acid (*P. bilaiae*). Exudation rates of oxalate decreased in response to lead exposure, while exudation rates of citrate were less affected. Total production under poor nutrient conditions was low, except for *A. niger*, for which no significant difference was found between the poor and rich control. Maximum exudation rates were 20 µmol oxalic acid g^{-1} biomass h^{-1} (*A. niger*) and 20 µmol citric acid g^{-1} biomass h^{-1} (*P. bilaiae*), in the presence of the contaminated soil, but only 5 µmol organic acids g^{-1} biomass h^{-1}, in total, for the *Penicillium* sp. There was a significant mobilization of metals from the soils in the carbon rich treatments and maximum release of Pb was 12% from the soils after 5 days. This was not sufficient to bring down the remaining concentration to the target level 300 mg kg^{-1} from initial levels of 3,800, 1,600, and 370 mg kg^{-1}in the three soils. Target levels for Ni, Zn, and Cu, were 120, 500, and 200 mg kg^{-1}, respectively, and were prior to the bioremediation already below these concentrations (except for Cu Soil 1). However, maximum release of Ni, Zn, and Cu was 28%, 35%, and 90%, respectively. The release of metals was related to the production of chelating acids, but also to the pH-decrease. This illustrates the potential to use fungi exudates in bioremediation of contaminated soil. Nonetheless, the extent of the generation of organic acids is depending on several processes and mechanisms that need to be further investigated.

Keywords: Bioremediation - Citric acid - Fungi - Lead - Organic acids - Oxalic acid

Wan Azlina Ahmad[a], Zainul Akmar Zakaria[a], Ali Reza Khasim[a], Muhamad Anuar Alias[a] and Shaik Muhammad Hasbullah Shaik Ismail[a]. ([a]Department of Chemistry, Faculty of Science, Universiti Teknologi Malaysia, 81310 Skudai, Johor, Malaysia). **Pilot-scale removal of chromium from industrial wastewater using the ChromeBac™ system. Bioresource Technology, Volume 101(12) (2010): 4371-4378**

The enzymatic reduction of Cr(VI) to Cr(III) by Cr(VI) resistant bacteria followed by chemical precipitation constitutes the ChromeBac™ system. *Acinetobacter haemolyticus* was immobilized onto carrier material inside a 0.2 m^3 bioreactor. Neutralized electroplating wastewater with Cr(VI) concentration of 17–81 mg L^{-1} was fed into the bioreactor (0.11–0.33 m^3 h^{-1}). Complete Cr(VI) reduction to Cr(III) was obtained immediately after the start of bioreactor operation. Together with the flocculation, coagulation and filtration, outflow concentration of less than 0.02 mg Cr(VI) L^{-1} and 1 mg total Cr L^{-1} were always obtained. Performance of the bioreactor

was not affected by fluctuations in pH (6.2–8.4), Cr(VI) (17–81 mg L^{-1}), nutrient (liquid pineapple waste, 1–20% v/v) and temperature (30–38 °C). Standby periods of up to 10 days can be tolerated without loss in activity. A robust yet effective biotechnology to remove chromium from wastewater is thus demonstrated.

Keywords: Detoxification; Electroplating; *Acinetobacter*; Cr(VI); Pineapple wastewater

Tekin Şahan[a], Hasan Ceylan[b], Nurettin Şahiner[c] and Nahit Aktaş[a]. ([a]Yuzuncu Yil University, Faculty of Engineering and Architecture, Chemical Engineering Department, 65080 Van, Turkey. [b] Yuzuncu Yil University, Faculty of Science and Art, Chemistry Department, 65080 Van, Turkey. [c] Canakkale Onsekiz Mart University, Faculty of Science and Art, Chemistry Department, 17020 Canakkale, Turkey). **Optimization of removal conditions of copper ions from aqueous solutions by *Trametes versicolor*. Bioresource Technology, Volume 101(12) (2010): 4520-4526**

A multi-step response surface methodology was successfully applied to optimize the biosorption conditions for the maximum removal of Cu(II) ions from aqueous solutions using *Trametes versicolor* fungi as a biosorbent. In the first step, the most effective medium factors, which are pH, temperature and initial Cu(II) concentration, on biosorption of Cu(II), were determined through Plackett–Burman Design. Then steepest accent followed by central composite design steps were utilized to evaluate the optimum biosorption conditions for the maximum Cu(II) ions removal. Based on the statistic analysis; the optimum conditions were obtained 5.51, 20.13 °C and 60.98 mg/L as medium pH, medium temperature and initial Cu(II) concentration, respectively. Finally the analysis of variance (ANOVA) of central composite design showed the proposed quadratic model fitted experimental data very well.

Keywords: Biosorption; Copper ion; Heavy metal removal; Response surface methodology; *Trametes versicolor*

Jinshao Ye[a, b], Hua Yin[a], Bixian Mai[b], Hui Peng[a], Huaming Qin[a], Baoyan He[a] and Na Zhang[a]. ([a] Department of Environmental Engineering, Jinan University, Guangzhou 510632, PR China. [b] State Key Laboratory of Organic Geochemistry, Guangzhou Institute of Geochemistry, Chinese Academy of Science, Guangzhou 510640, PR China). **Biosorption of chromium from aqueous solution and electroplating wastewater using mixture of *Candida lipolytica* and dewatered sewage sludge. Bioresource Technology, Volume 101 (11) (2010): 3893-3902**

In this study, the objective was to investigate Cr removal from aqueous solutions, as well as Cr, Cu, Ni and Zn from electroplating wastewaters by the mixture of *Candida lipolytica* and sewage sludge. The bioreduction ratios of Cr(VI) and the removal ratios of total Cr showed that initial pH, biosorbent dosage and contact time were the important parameters for Cr biosorption. The range of optimal pH for the mixture (1–5) was wider than *C. lipolytica* (1–4) and sewage sludge (2–4), respectively. Biosorption and bioreduction potentials of living *C. lipolytica* were better than those of cell wall and cytoplasm. Bonded hydroxyl group, CH_2 asymmetric stretch, amide I, amide II, amide III, secondary amide, pyridine(I)β(C–H) and pyridine(II)β(C–H) were detected in the biosorbent and they were the functional groups for binding Cr. The effect of Cu and Zn in combination was significant on the removal of total Cr and the bioreduction of Cr(VI).

Keywords: *Candida lipolytica*; Chromium; Sewage sludge; Biosorption; Heavy metal

Hector Monclús[a], Jan Sipma[a], Giuliana Ferrero[a], Ignasi Rodriguez-Roda[a, b] and Joaquim Comas[a]. ([a] Laboratory of Chemical and Environmental Engineering (LEQUiA), Institute of the Environment, University of Girona, E17071 Girona, Spain. [b] Catalan Institute for Water Research (ICRA), Scientific and Technological Park of the University of Girona, H_2O Building, Emili Grahit 101, 17003 Girona, Spain). Biological nutrient removal in an MBR treating municipal wastewater with special focus on biological phosphorus removal. Bioresource Technology, Volume 101 (11) (2010): 3984-3991

The performance of an MBR pilot plant for biological nutrient removal was evaluated during 210 days of operation. The set point values for the internal recycles were determined in advance with the use of an optimisation spreadsheet based on the ASM2d model to optimise the simultaneous removal of C, N and P. The biological nutrient removal (BNR) efficiencies were high from the start of operation with COD and N removal efficiencies of 92 ± 6% and 89 ± 7, respectively. During the course of the experiment P removal efficiencies increased and finally a P-removal efficiency of 92% was achieved. The activity of poly-phosphate accumulating organisms (PAOs) and denitrifying poly-phosphate accumulating organisms (DPAOs) increased and the specific phosphate accumulation rates after 150 days of operation amounted to 13.6 mg P g^{-1}VSS h^{-1} and 5.6 mg P g^{-1}VSS h^{-1}, for PAOs and DPAOs, respectively.

Keywords: Biological nutrient removal (BNR); Enhanced biological phosphorus removal (EBPR); Membrane bioreactor (MBR); Poly-phosphate accumulating organisms (PAOs); UCT configuration

Ying Teng[a], Yongming Luo[a], Mingming Sun[a], Zengjun Liu[a], Zhengao Li[a] and Peter Christie[b]. ([a] Key Laboratory of Soil Environment and Pollution Remediation, Institute of Soil Science, Chinese Academy of Sciences, Nanjing 210008, China. [b] Agri-Environment Branch, Agri-Food and Biosciences Institute, Newforge Lane, Belfast BT9 5PX, United Kingdom). Bioresource Technology, Volume 101 (11) (2010): 3984-3991

A microcosm study was conducted to test the bioremediation potential of *Paracoccus* sp. strain HPD-2 on an aged PAH-contaminated soil. Bioaugmented microcosms showed a 23.2% decrease in soil total PAH concentrations after 28 days, with a decline in average concentration from 9942 to 7638 μg kg^{-1} dry soil. The percentage degradation of 3-, 4- and 5(+6)-ring PAHs was 35.1%, 20.7% and 24.3%, respectively. Higher counts of culturable PAH-degrading bacteria, microbial biomass and enzyme activities were observed in bioaugmented soil. The bioaugmented microcosms showed significant increases ($p < 0.05$) in the average well-color development (AWCD) obtained by the BIOLOG ecoplate assay and Shannon–Weaver index (H) compared to the controls. Principal component analysis of BIOLOG data clearly differentiated between the bioaugmented and control microcosms, implying that bioaugmentation restored the microbiological functioning of the PAH-contaminated soil. The results suggest that bioaugmentation by *Paracoccus* sp. strain HPD-2 may be a promising bioremediation strategy for aged PAH-contaminated soils.

Keywords: Polycyclic aromatic hydrocarbons; Bioaugmentation; *Paracoccus* sp. strain HPD-2; Biodegradation; Microbial activities

Francesca Pagnanelli[a], Carolina Cruz Viggi[a] and Luigi Toro[a]. ([a] Department of Chemistry, Sapienza University of Rome, P.le Aldo Moro 5, 00185 Rome, Italy). **Isolation and quantification of cadmium removal mechanisms in batch reactors inoculated by sulphate reducing bacteria: Biosorption versus bioprecipitation. Bioresource Technology, Volume 101 (9) (2010): 2981-2987**

Biosorbing properties of sulphate reducing bacteria were tested to distinguish the amount of cadmium removed by bioprecipitation from that bound onto biomass surface (biosorption). Experimental results of cadmium abatement in batch growth tests (bioprecipitation tests) were then compared with metabolism-independent binding properties of SRB cell wall surface (biosorption tests performed with dead biomass). Experimental results showed that SRB inoculum removed 59 ± 5% of sulphates in 21 days even in presence of cadmium (0–36 mmol L^{-1}), while non-monotonous kinetic effects were observed for increasing Cd concentrations. Comparison between bioprecipitation and biosorption tests denoted a significant contribution of biosorption (77%) in total Cd removal (0.40 ± 0.01 mmol g^{-1}). Characterisation of bacterial acid–base surface properties by potentiometric titrations and mechanistic modelling denoted that carboxylic, phosphate and amino groups of cell wall are the main responsible of metal removal by biosorption mechanism.

Jung-Chun Chen[a], Kai-Sung Wang[b, c], Hsien Chen[b, c], Chi-Yuan Lu[b, c], Lung-Chiu Huang[b, c], Heng-Ching Li[b, c], Tzu-Huan Peng[b, c] and Shih-Hsien Chang[b, c]. ([a] Department of Soil and Water Conservation, National Chung-Hsing University, Taichung 402, Taiwan, ROC. [b] Department of Public Health, Chung-Shan Medical University, Taichung 402, Taiwan, ROC. [c] Department of Family and Community Medicine, Chung Shan Medical University Hospital, Taichung 402, Taiwan, ROC). **Phytoremediation of Cr(III) by *Ipomonea aquatica* (water spinach) from water in the presence of EDTA and chloride: Effects of Cr speciation. Bioresource Technology, Volume 101 (9) (2010): 3033-3039**

Wastewater is often co-contaminated with chromium, chelating agents, and chloride. Influences of Cr^{3+} speciation on Cr phytoremediation by *Ipomonea aquatica* were investigated. MINEQL+ was employed to estimate Cr speciation. Statistic regression was used to investigate the relationships between Cr speciation and accumulation. *I. aquatica* accumulated high Cr concentration (13,217 mg kg^{-1}) in the root at Cr^{3+} of 10 mg l^{-1} and EDTA of 10^{-4} M after 14 d growth. Pearson correlation analysis indicates that root Cr concentration significantly correlated with Cr–EDTA speciation ($r = 0.67$, $p < 0.05$) and Cr–Cl speciation ($r = 0.91$, $p < 0.01$). Shoot Cr concentration also significantly correlated with Cr–Cl speciation ($r = 0.97$, $p < 0.01$). An increase in Cl^- concentration to 1.72×10^{-4} M enhanced root Cr concentration; however, the accumulation of root Cr was inhibited at high Cl^- concentration (5.76×10^{-5} M). Microscopic image showed that a high portion of Cr^{3+} accumulated on the root surface.

Keywords: Phytoremediation; Macrophyte; MINEQL+; Speciation

Antonella Anastasi[a], Federica Spina[a], Valeria Prigione[a], Valeria Tigini[a], Pietro Giansanti[b] and Giovanna Cristina Varese[a]. ([a] Dipartimento di Biologia Vegetale, Università degli Studi di Torino, Viale Mattioli 25, 10125 Torino, Italy. [b] Arpa Piemonte, Dipartimento di Torino, Laboratorio di Ecotossicologia e Mutagenesi, Via Sabaudia, 164, 10095 Grugliasco

(TO), Italy). Scale-up of a bioprocess for textile wastewater treatment using *Bjerkandera adusta*. Bioresource Technology, Volume 101 (9) (2010): 3067-3075

Twelve basidiomycetes were investigated for their ability to degrade 13 industrial dyes and to treat four model wastewaters from textile and tannery industry, defined on the basis of discharged amounts, economic relevance and representativeness of chemical structures of the contained dyes. The best degradation yields were recorded for one strain of *Bjerkandera adusta* able to completely decolourise most of the dyes and to decolourise and detoxify three simulated wastewaters, showing a significant physiological versatility which is very useful for application purposes. The effects of different nutrient sources were investigated in order to optimize the yields of decolourisation and detoxification. Manganese-peroxidase and manganese-independent peroxidase were the only recorded enzymatic activities. In order to evaluate its true bioremediation potential, this strain was packed in a fixed-bed bioreactor, for treatment of large volumes of a real wastewater. The fungus resulted effective during 10 cycles of decolourisation, remaining active for a very long period, in non-sterile conditions.

Keywords: Biodegradation; Bioreactor; Ecotoxicity; Industrial dyes; White-rot fungi

Sushanta Kumar Saha[a, b], Palanisami Swaminathan[a], C. Raghavan[a, c], Lakshmanan Uma[a] and Gopalakrishnan Subramanian[a]. ([a] National Facility for Marine Cyanobacteria, Bharathidasan University, Tiruchirappalli 620024, India. [b] Division of Biological Sciences, Molecular Biology Section, University of California, San Diego, La Jolla, CA 92093, USA. [c] Institute of Microbiology and Immunology, University Clinic of Ulm, Ulm D89081, Germany). **Ligninolytic and antioxidative enzymes of a marine cyanobacterium *Oscillatoria willei* BDU 130511 during Poly R-478 decolourization. Bioresource Technology, Volume 101 (9) (2010): 3076-3084**

Removal of combined nitrogen and addition of Poly R-478 to the growth medium enhanced oxidative stress, and altered the activities of ligninolytic enzymes of *Oscillatoria willei* BDU 130511. The activities of ligninolytic and antioxidative enzymes (LiP-like, LAC, PPO, SOD, POD, CAT, and APX) were increased upon nitrogen limitation and dye supplementation. The metabolic enzymes tested (GR, GPX, EST, and MDH) showed differential expressions under varied growth conditions. Up on nitrogen limitation, *O. willei* BDU 130511 showed enhanced ligninolytic activity as shown by α-keto-γ-methylthiolbutyric acid (KTBA) oxidation and increased H_2O_2 production. The organism decolourized 52% of Poly R-478 due to partial degradation and adsorption of dye particles from dye-added medium after 7 days of growth. This manuscript discusses the responses of ligninolytic and antioxidative enzymes of *O. willei* BDU 130511 during Poly R-478 decolourization/degradation, and the organism's potential in bioremediation.

Keywords: Antioxidative; Decolourization; Ligninolytic; *Oscillatoria willei*; Poly R-478

Abbreviations: ABTS, 2,2′-azino-bis(3-ethylbenzthiazoline-6-sulfonate); DAB, 3,3′-diaminobenzidine tetrahydrochloride; DOPA, 3,4-dihydroxy phenylalanine; KTBA, α-keto-γ-methylthiolbutyric acid; NAD, nicotinamide adenine dinucleotide; NBT, nitroblue tetrazolium chloride; PMS, *n*-methylphenazonium methosulphate; APX, ascorbate peroxidase; CAT, catalase; EST, esterase; GPX, glutathione peroxidase; GR, glutathione reductase; LAC, laccase; LiP, lignin peroxidase; MDH, malate dehydrogenase; POD, peroxidase; PPO, polyphenol oxidase; SOD, superoxide dismutase

Mang Lu[a], Zhongzhi Zhang[a], Wei Qiao[a], Xiaofang Wei[a, b], Yueming Guan[a], Qingxia Ma[a] and Yingchun Guan[a]. ([a] State Key Laboratory of Heavy Oil Processing, China University of Petroleum, Beijing 102249, China. [b] Research Institute of Petroleum Exploration and Development, Petrochina, Beijing 100083, China). Remediation of petroleum-contaminated soil after composting by sequential treatment with Fenton-like oxidation and biodegradation. Bioresource Technology, Volume 101 (7) (2010): 2106-2113

A laboratory study was conducted to enhance removal of residual contaminants after composting in a highly petroleum-contaminated soil by combining Fenton-like pretreatment with biodegradation. The contaminants were characterized by Fourier transform ion cyclotron resonance mass spectrometry (FT-ICR MS) during soil treatment. The optimum molar ratio of H_2O_2 and Fe^{3+} was 300/1 determined in batch experiments. At the end of Fenton-like treatment, total dichloromethane-extractable organics (TEO) decreased from 32,400 to 21,800 mg kg^{-1} soil, and the toxicity of soil was reduced greatly in the preoxidation process. A significant loss of the number of soil microorganisms was observed in the Fenton-like reaction. During the microbial treatment period, 50.6% of TEO was destroyed. Numerous varieties of polar compounds containing nitrogen and oxygen were identified by FT-ICR MS. The number of compounds containing two oxygen atoms dropped from 604 to 163 during Fenton-like oxidation, and increased again to 577 after biodegradation.

Keywords: Bioremediation; Bioslurry; Fourier transform ion cyclotron resonance mass spectrometry; The van Krevelen diagram

K. Tsekova[a], D. Todorova[a], V. Dencheva[a] and S. Ganeva[b]. ([a] Microbial Ecology Department, Institute of Microbiology, Bulgarian Academy of Sciences, "Acad. G. Bonchev" Str., bl. 26, 1113 Sofia, Bulgaria. [b] Faculty of Chemistry, Sofia University, Sofia, Bulgaria). Biosorption of copper(II) and cadmium(II) from aqueous solutions by free and immobilized biomass of *Aspergillus niger*. Bioresource Technology, Volume 101 (6) (2010): 1727-1731

This study investigates the ability of *Aspergillus niger* resting cells entrapped into poly(vinyl alcohol) (PVA) network to remove Cu(II) and Cd(II) from single ions solutions. The performance of free and immobilized biosorbent was evaluated by equilibrium and kinetic studies. The PVA-immobilized fungal biosorbent removed Cu(II) and Cd(II) rapidly and efficiently with maximum metal removal capacities of 34.13 mg/g and 60.24 mg/g, respectively. These values of heavy metal uptake at equilibrium were higher than the amount of Cu(II) and Cd(II) removal by free biomass (17.60 mg/g and 69.44 mg/g, respectively). Biosorption equilibrium data were best described by Langmuir isotherm model. The biosorption kinetics followed the pseudo-second order model and intraparticle diffusion equation. The results obtained suggest that the immobilized biosorbent holds great potential for wastewater treatment applications.

Keywords: *Aspergillus niger*; Immobilized cells biosorption; Copper; Cadmium

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Industrial Research, Lahore, Pakistan). Removal of Pb(II) and Cd(II) from water by adsorption on peels of banana. Bioresource Technology, Volume 101 (6) (2010): 1752-1755

The adsorption of lead(II) and cadmium(II) on peels of banana has been studied in batch mode using flame atomic absorption spectroscopy for metal estimation. Concerned parameters like adsorbent dose, pH, contact time and agitation speed were investigated. Langmuir, Freundlich and Temkin isotherms were employed to describe adsorption equilibrium. The maximum amounts of cadmium(II) and lead(II) adsorbed (*qm*), as evaluated by Langmuir isotherm, were 5.71 mg and 2.18 mg per gram of powder of banana peels, respectively. Study concluded that banana peels, a waste material, have good potential as an adsorbent to remove toxic metals like lead and cadmium from water.

Keywords: Langmuir isotherm; Freundlich isotherm; Temkin isotherm; Adsorption; Utilization of banana peels

Brent C. Christner[1]. ([1]Department of Biological Sciences, Louisiana State University, Baton Rouge, LA 70803, USA). Bioprospecting for microbial products that affect ice crystal formation and growth. Applied Microbiology and Biotechnology, Volume 85 (3) (2010): 481-489

At low temperatures, some organisms produce proteins that affect ice nucleation, ice crystal structure, and/or the process of recrystallization. Based on their ice-interacting properties, these proteins provide an advantage to species that commonly experience the phase change from water to ice or rarely experience temperatures above the melting point. Substances that bind, inhibit or enhance, and control the size, shape, and growth of ice crystals could offer new possibilities for a number of agricultural, biomedical, and industrial applications. Since their discovery more than 40 years ago, ice nucleating and structuring proteins have been used in cryopreservation, frozen food preparation, transgenic crops, and even weather modification. Ice-interacting proteins have demonstrated commercial value in industrial applications; however, the full biotechnological potential of these products has yet to be fully realized. The Earth's cold biosphere contains an almost endless diversity of microorganisms to bioprospect for microbial compounds with novel ice-interacting properties. Microorganisms are the most appropriate biochemical factories to cost effectively produce ice nucleating and structuring proteins on large commercial scales.

Keywords: Ice-nucleating proteins - Antifreeze proteins - Ice-structuring proteins - Cryopreservation - Bioprospecting - Cryosphere

Abdul Rehman[1], S. Awais Butt and Shahida Hasnain. ([1]Department of Microbiology and Molecular Genetics, University of the Punjab, New Campus, Lahore 54590, Pakistan). Isolation and characterization of arsenite oxidizing *Pseudomonas lubricans* and its potential use in bioremediation of wastewater. African Journal of Biotechnology, Vol. 9 (10) (2010): 1493-1498

A bacterium, *Pseudomonas lubricans*, isolated from heavy metal laden industrial wastewater, has been shown to tolerate multiple heavy metals suggesting its importance in bioremediation of industrial effluents. *P. lubricans* tolerated As(III) up to 3 mg ml^{-1}, Cu^{2+} up to 0.7 mg ml^{-1}, Hg^{2+} up to 0.4 mg ml^{-1}, Ni^{2+} up to 0.4 mg ml^{-1} and Cr^{6+} up to 0.5 mg ml^{-1}. *P. lubricans* showed optimum growth at pH 7 while optimum temperature for growth was 30°C. *P. lubricans* could oxidize As(III) 42% (42 µg mg^{-1} of protein), 78% (78 µg mg^{-1} of protein) and 95% (95 µg mg^{-1}

of protein) from the medium after 24, 48 and 72 h of incubation at optimal conditions, respectively. The arsenite oxidizing ability shown by *P. lubricans* indicates its potential application in biological treatment of wastewaters contaminated with arsenic.

Abbreviations: LB, Luria-bertani; **MIC,** minimum inhibitory concentration; **DCPIP,** dichlorophenolindophenol; **PMS,** phenazine methosilfate.

Key words: Arsenic, wastewater, *Pseudomonas lubricans*, bioremediation.

G. Wultsch[1], D. Haas[2], H. Galler[2], G. Feierl[2], A. Melkes[2] and F. F. Reinthaler[2]. ([1]Occupational Medicine, AMEZ Graz, Raiffeisenstraße 60, 8010 Graz, Austria, [2]Institute of Hygiene, Microbiology and Environmental Medicine, Medical University Graz, Universitätsplatz 4, 8010 Graz, Austria). **Bioaerosol emissions in a poultry litter burning plant. Aerobiologia, Volume 26 (1) (2010): 63-73**

This study investigates the exposure of workers to biological particles in a poultry litter burning plant in operation. The microorganism concentrations were examined at different workplaces during procedures leading to increased emissions. The concentrations of culturable airborne mesophilic, xerophilic and thermophilic microorganisms in the ambient air were tested inside and outside of the burning plant using two different methods of measuring. The focus of this study was on the quantitative evaluation of culturable bacteria as well as the quantitative and qualitative evaluation of gram-negative bacteria, fungi and thermophilic actinomycetes. The maximum airborne concentrations were found in the delivery hall. Mesophilic bacteria concentrations reached up to 1.7×10^6 CFU/m^3; gram-negative bacteria up to 9.1×10^2 CFU/m^3. Fungal propagule concentrations for xerophilic fungi were between 1.2×10^3 and 2.9×10^4 CFU/m^3 and for mesophilic fungi between 4.4×10^2 and 2.9×10^4 CFU/m^3. Among fungi, *Aspergillus niger*, *Eurotium herbariorum* and *Scopulariopsis brevicaulis* species were dominant. Thermophilic actinomycetes reached airborne concentrations of 8.7×10^4 CFU/m^3, with increased concentrations of the pathogens causing extrinsic allergic alveolitis. The high concentrations of airborne microorganisms in poultry litter burning plants and the potential hazard of the intake of microorganisms including potential pathogens require the introduction of consistent measures in both technical areas and personnel management.

Keywords: Poultry litter - Bioaerosol - CFU - Fungi - Bacteria

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Microorganisms produce biosurfactant (BS)/bioemulsifier (BE) with wide structural and functional diversity which consequently results in the adoption of different techniques to investigate these diverse amphiphilic molecules. This review aims to compile information on

different microbial screening methods, surface active products extraction procedures, and analytical terminologies used in this field. Different methods for screening microbial culture broth or cell biomass for surface active compounds production are also presented and their possible advantages and disadvantages highlighted. In addition, the most common methods for purification, detection, and structure determination for a wide range of BS and BE are introduced. Simple techniques such as precipitation using acetone, ammonium sulphate, solvent extraction, ultrafiltration, ion exchange, dialysis, ultrafiltration, lyophilization, isoelectric focusing (IEF), and thin layer chromatography (TLC) are described. Other more elaborate techniques including high pressure liquid chromatography (HPLC), infra red (IR), gas chromatography-mass spectroscopy (GC-MS), nuclear magnetic resonance (NMR), and fast atom bombardment mass spectroscopy (FAB-MS), protein digestion and amino acid sequencing are also elucidated. Various experimental strategies including static light scattering and hydrodynamic characterization for micelles have been discussed. A combination of various analytical methods are often essential in this area of research and a numbers of trials and errors to isolate, purify and characterize various surface active agents are required. This review introduces the various methodologies that are indispensable for studying biosurfactants and bioemulsifiers

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Heavy metal pollution of soil is a significant environmental problem with a negative potential impact on human health and agriculture. Rhizosphere, as an important interface of soil and plants, plays a significant role in phytoremediation of contaminated soil by heavy metals, in which, microbial populations are known to affect heavy metal mobility and availability to the plant through release of chelating agents, acidification, phosphate solubilization and redox changes, and therefore, have potential to enhance phytoremediation processes. Phytoremediation strategies with appropriate heavy metal-adapted rhizobacteria or mycorrhizas have received more and more attention. In addition, some plants possess a range of potential mechanisms that may be involved in the detoxification of heavy metals, and they manage to survive under metal stresses. High tolerance to heavy metal toxicity could rely either on reduced uptake or increased plant internal sequestration, which is manifested by an interaction between a genotype and its environment.A coordinated network of molecular processes provides plants with multiple metal-detoxifying mechanisms and repair capabilities. The growing application of molecular genetic technologies has led to an increased understanding of mechanisms of heavy metal tolerance/accumulation in plants and, subsequently, many transgenic plants with increased heavy metal resistance, as well as increased uptake of heavy metals, have been developed for the purpose of phytoremediation. This article reviews advantages, possible mechanisms, current status and future direction of phytoremediation for heavy-metal–contaminated soils.

P. C. Suryawanshi[1], A. B. Chaudhari[2], R. M. Kothari[1]. ([1]Jain R & D Laboratory, Agri Park, Jain Hills, Jain Irrigation Systems Ltd, Jalgaon, India. [2]Department of Microbiology, School of Life Sciences, North Maharashtra University, Jalgaon, India). Thermophilic anaerobic digestion: the best option for waste treatment. Critical Reviews in Biotechnology, Vol. 30 (1) (2010): 70-77

After introducing thermophilic anaerobic digestion (AD), characteristics of thermophilic methanogens are provided. Accordingly, (a) site of occurrence, (b) morphological characteristics (shape and motility), (c) biochemical characteristics (Gram character and % G+C profile), (d) nutritional characteristics (NaCl requirement and substrate specificity), and (e) growth characteristics (pH and temperature) of thermophilic methanogens are described. Some studies of the thermophilic AD are cited with their operational management problems. Subsequently, strategies to maximize net energy production are given, including mode of heating the bioreactors, role of agitation to promote AD performance and mode/intensity of mixing. Finally, advantages as well as drawbacks of AD under thermophilic conditions are given, concluding with its applications.

Dawen Gao[1,2], Lina Du[1], Jiaoling Yang[1], Wei-Min Wu[3], Hong Liang[1]. ([1]School of Forestry, Northeast Forestry University, Harbin, P.R. China. [2]State Key Laboratory of Urban Water Resource and Environment, Harbin Institute of Technology, Harbin, P.R. China. [3]Department of Civil and Environmental Engineering, Stanford University, Stanford, CA, USA). A critical review of the application of white rot fungus to environmental pollution control.

Research on white rot fungi for environmental biotechnology has been conducted for more than 20 years. In this article, we have reviewed processes for cell growth and enzyme production including the factors influencing enzyme productivity and the methods for enhancement of enzyme production. Significant progress has been achieved in molecular biology related to white rot fungi, especially related to the extraction of genetic material (RNA and DNA), gene cloning and the construction of genetically engineered microorganisms. The development of biotechnologies using white rot fungi for environmental pollution control has been implemented to treat various refractory wastes and to bioremediate contaminated soils. The current status and future research needs for fundamentals and application are addressed in this review.

Sheikh M. Basha[1], Hifza Mazhar[1] and Hemanth K. N. Vasanthaiah[1]. ([1]Division of Plant Biotechnology, Center for Viticulture and Small Fruit Research, Florida A & M University, 6505 Mahan Drive, Tallahassee, FL 32317, USA). Proteomics Approach to Identify Unique Xylem Sap Proteins in Pierce's Disease-Tolerant *Vitis* Species. Applied Biochemistry and Biotechnology, Volume 160 (3) (2010): 932-944

Pierce's disease (PD) is a destructive bacterial disease of grapes caused by *Xylella fastidiosa* which is xylem-confined. The tolerance level to this disease varies among *Vitis* species. Our research was aimed at identifying unique xylem sap proteins present in PD-tolerant *Vitis* species. The results showed wide variation in the xylem sap protein composition, where a set of polypeptides with p*I* between 4.5 and 4.7 and M_r of 31 kDa were present in abundant amount in muscadine (*Vitis rotundifolia*, PD-tolerant), in reduced levels in Florida hybrid bunch (*Vitis* spp., PD-tolerant) and absent in bunch grapes (*Vitis vinifera*, PD-susceptible). Liquid

chromatography/mass spectrometry/mass spectrometry analysis of these proteins revealed their similarity to β-1, 3-glucanase, peroxidase, and a subunit of oxygen-evolving enhancer protein 1, which are known to play role in defense and oxygen generation. In addition, the amount of free amino acids and soluble sugars was found to be significantly lower in xylem sap of muscadine genotypes compared to *V. vinifera* genotypes, indicating that the higher nutritional value of bunch grape sap may be more suitable for *Xylella* growth. These data suggest that the presence of these unique proteins in xylem sap is vital for PD tolerance in muscadine and Florida hybrid bunch grapes.

Keywords: Grape - Pierce's disease - Differential protein expression - Xylem sap

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The removal of metal ions from aqueous solutions by biosorption plays an important role in water pollution control. In this study, dried leaves and branches of poplar trees were studied for removing some toxic elements (cadmium, lead, and uranium) from aqueous solutions. The equilibrium experiments were systematically carried out in a batch process, covering various process parameters that include agitation time, adsorbent size and dosage, initial cadmium, lead and uranium concentration, and pH of the aqueous solution. Adsorption behavior was found to follow Freundlich and Langmuir isotherms. The results have shown that both dried leaves and branches can be effectively used for removing uranium, while only branches were found to remove lead and cadmium completely from the aqueous solution. The maximum biosorption capacity of leaves for uranium was found to be 2.3 mg g^{-1} and 1.7 mg g^{-1} and 2.1 mg g^{-1} for lead and cadmium on branches, respectively. In addition, the studied biomass materials were used in removing lead and cadmium from contaminated water and the method was found to be effective.

Keywords: Poplar - Uranium - Cadmium - Lead - Biosorption - Wastewater - Pollution

G. Ofori-Sarpong[a], M. Tien[b] and K. Osseo-Asare[a, c]. ([a] Dept. of Energy and Mineral Engineering, Penn State University, University Park, PA 16802, USA. [b] Dept. of Biochemistry and Molecular Biology, Penn State University, University Park, PA 16802, USA. [c] Dept. of Materials Science and Engineering, Penn State University, University Park, PA 16802, USA). Myco-hydrometallurgy: Coal model for potential reduction of preg-robbing capacity of carbonaceous gold ores using the fungus, *Phanerochaete chrysosporium.* Hydrometallurgy, Volume 102 (1-4) (2010): 66-72

During the cyanidation process for gold extraction, natural carbonaceous matter (CM) present in refractory gold ores adsorbs dissolved aurocyanide complexes, thereby reducing gold extraction — a phenomenon known as preg-robbing. The natural CM consists mainly of elemental carbon, humic acids and hydrocarbons, and coal can be used to model the behavior of CM. In this study, samples of lignite, sub-bituminous, bituminous and anthracite coals were used as surrogate materials to test the capability of the fungus *Phanerochaete chrysosporium* to reduce the preg-robbing capacity of CM.

By utilizing several growth media including glucose, millet and wheat bran it was established that millet and wheat bran provided the best environment for fungal growth and biomodification of coal. As-received, control and bio-treated coal samples were subjected to gold adsorption tests and the results indicate that *P. chrysosporium* can decrease the gold adsorption ability of coal by over 90% depending on the growth media used. Reduction in gold adsorption was more pronounced in anthracite due to its high ability to adsorb gold in the as-received form. The results demonstrate a potentially effective alternative pretreatment process that utilizes the fungus, *P. chrysosporium,* in reducing preg-robbing in gold extraction.

Keywords: Myco-hydrometallurgy; Fungi; *Phanerochaete chrysosporium*; Biomodification; Coals; Carbonaceous matter; Preg-robbing; Gold adsorption; Cyanidation

Biotransformation

Richard W. Eaton[1] and Peter Sandusky[2]. ([1]Southern Regional Research Center, Agricultural Research Service, US Department of Agriculture, 1100 Robert E. Lee Blvd., New Orleans, LA 70124, USA, [2]Department of Environmental Science and Engineering, The University of North Carolina, Chapel Hill, NC 27599, USA). **Biotransformations of (+/−)-geosmin by terpene-degrading bacteria. Biodegradation, Volume 21(1) (2010): 71-79**

Two terpene-degrading bacteria able to transform (+/−)-geosmin have been identified. *Pseudomonas* sp. SBR3-tpnb, following growth on γ-terpinene, converts (+/−)-geosmin to several products; the major products are ketogeosmins. *Rhodococcus wratislaviensis* DLC-cam, isolated on D-camphor, also converts (+/−)-geosmin to several oxidation products, primarily ketogeosmins identical to those produced by strain SBR3-tpnb as well as hydroxygeosmins. This conversion appears to be inducible by (+/−)-geosmin and not by D-camphor.

Keywords: Geosmin - Biotransformation - Terpene - *Pseudomonas* - *Rhodococcus wratislaviensis*

Priti N. Chaudhari[1], Bhushan L. Chaudhari[1] and Sudhir B. Chincholkar[1] ([1]School of Life Sciences, North Maharashtra University, P B 80, Jalgaon, 425 001, India). **Cholesterol biotransformation to androsta-1,4-diene-3,17-dione by growing cells of *Chryseobacterium gleum.* Biotechnology Letters, Volume 32(5) (2010): 695-699**

Cholesterol oxidase activity was studied during biotransformation of cholesterol to androsta-1,4-diene-3,17-dione (ADD) by *Chryseobacterium gleum.* Spent LB media, containing cholesterol (3 mM≈1 g l^{-1}) where the bacterium was grown for 24 h, at 30°C with constant shaking at 120 rpm, had the highest enzyme activity (167 U mg^{-1}). The growing cells produced 0.076 g ADD from 1 g cholesterol l^{-1}.

Keywords: Androsta-1,4-diene-3,17-dione - Biotransformation - Cholesterol - *Chryseobacterium gleum*

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Dublin 4, Ireland). Biodegradation and biotransformation of organofluorine compounds. Biotechnology Letters, Volume 32(3) (2010): 351-359

The carbon–fluorine bond is one of the strongest in nature, and the increasing use of organofluorine compounds in agriculture, human and veterinary medicine, and industry has raised concerns about their fate in the environment. Microorganisms can degrade organofluorine compounds, either via specific enzymatic hydrolysis of the C–F bond, or through transformation by catabolic enzymes with broad substrate specificities. Here our current understanding of organofluorine catabolism in microorganisms is summarised.

Keywords: Bioremediation - Dehalogenase - Fluoride - Fluoroaromatic - Organofluoro compounds

Keshetty Srisailam[1] and Ciddi Veeresham[1]. ([1]niversity College of Pharmaceutical Sciences, Kakatiya University, Warangal, 506009, Andhra Pradesh, India). Biotransformation of Celecoxib Using Microbial Cultures. Applied Biochemistry and Biotechnology, Volume 160(7) (2010): 2075-2089

Microbial transformation studies can be used as models to simulate mammalian drug metabolism. In the present investigation, biotransformation of celecoxib was studied in microbial cultures. Bacterial, fungal, and yeast cultures were employed in the present study to elucidate the metabolism of celecoxib. The results indicate that a number of microorganisms metabolized celecoxib to various levels to yield eight metabolites, which were identified by high-performance liquid chromatography diode array detection and liquid chromatography tandem mass spectrometry analyses. HPLC analysis of biotransformed products indicated that majority of the metabolites are more polar than the substrate celecoxib. The major metabolite was found to be hydroxymethyl metabolite of celecoxib, while the remaining metabolites were produced by carboxylation, methylation, acetylation, or combination of these reactions. The methyl hydroxylation and further conversion to carboxylic acid was known to occur in metabolism by mammals. The results further support the use of microorganisms for simulating mammalian metabolism of drugs.

Keywords: Celecoxib - Biotransformation - HPLC - LC–MS/MS - Metabolite - Microorganisms

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The pharmaceutical diclofenac (DCF) is released in considerably high amounts to the aquatic environment. Photo-transformation of DCF was reported as the main degradation pathway in surface waters and was found to produce metabolites with enhanced toxicity to the green algae *Scenedesmus vacuolatus*. We identified and subsequently confirmed 2-[2-(chlorophenyl)amino]benzaldehyde (CPAB) as a transformation product with enhanced toxicity

using effect-directed analysis. The EC_{50} of CPAB (4.8 mg/L) was a factor of 10 lower than that for DCF (48.1 mg/L), due to the higher hydrophobicity of CPAB (log K_{ow} = 3.62) compared with DCF (log D_{ow} = 2.04) at pH 7.0.

Effect-directed analysis of irradiated diclofenac results in the identification of one photo-transformation product responsible for the enhanced toxicity to *Scenedesmus vacuolatus*.

Keywords: Diclofenac; 2-[2-(Chlorophenyl)amino]benzaldehyde; Effect-directed analysis; Photo-transformation; *Scenedesmus vacuolatus*; MODELKEY

Albert L. Juhasz[a], Euan Smith[a], Natasha Waller[b], Richard Stewart[c] and John Weber[a]. ([a]Centre for Environmental Risk Assessment and Remediation, University of South Australia, Mawson Lakes Campus, SA 5095, Australia, [b] CSIRO Land and Water, Glen Osmond, SA 5064, Australia, [c] Remediate, Kent Town, SA 5067, Australia). **Bioavailability of residual polycyclic aromatic hydrocarbons following enhanced natural attenuation of creosote-contaminated soil. Environmental Pollution, Volume 158(2) (2010):** 585-591

The impact of residual PAHs (2250 ± 71 µg total PAHs g^{-1}) following enhanced natural attenuation (ENA) of creosote-contaminated soil (7767 ± 1286 µg total PAHs g^{-1}) was assessed using a variety of ecological assays. Microtox™ results for aqueous soil extracts indicated that there was no significant difference in EC_{50} values for uncontaminated, pre- and post-remediated soil. However, in studies conducted with *Eisenia fetida*, PAH bioaccumulation was reduced by up to 6.5-fold as a result of ENA. Similarly, *Beta vulgaris* L. biomass yields were increased 2.1-fold following ENA of creosote-contaminated soil. While earthworm and plant assays indicated that PAH bioavailability was reduced following ENA, the residual PAH fraction still exerted toxicological impacts on both receptors. Results from this study highlight that residual PAHs following ENA (presumably non-bioavailable to bioremediation) may still be bioavailable to important receptor organisms such as earthworms and plants.

Residual PAHs in creosote-contaminated soil following enhanced natural attenuation impacted negatively on ecological receptors.

Keywords: Bioavailability; Bioremediation; Creosote; Enhanced natural attenuation; PAHs

Douglas I. Stewart[a], Ian T. Burke[b], Danielle V. Hughes-Berry[b] and Robert A. Whittleston[b]. ([a] School of Civil Engineering, University of Leeds, Leeds LS2 9JT, UK, [b] School of Earth and Environment, Institute of Geological Sciences, University of Leeds, Leeds LS2 9JT, UK). **Microbially mediated chromate reduction in soil contaminated by highly alkaline leachate from chromium containing waste. Ecological Engineering, Volume 36(2) (2010): 211-**221

This paper reports an investigation into the possible fate of Cr(VI) that is migrating downwards from a chromium ore processing residue (COPR) tip into the underlying soils. This waste was deposited at a site in the north of England more than 100 years ago and is currently a cause for environmental concern because groundwater emerging from the waste is alkaline, visibly yellow and has an elevated Cr(VI) concentration. Sandy clay from immediately beneath the waste (assumed to be the topsoil layer prior to waste tipping) contains between about 600 and

3000 mg kg^{-1} of Cr, and around 60% of 0.5 N HCl extractable iron was present as reduced Fe(II). DNA fragments from soil bacteria were extracted from this soil, and microcosm experiments where the pH was adjusted to more neutral values showed that it contains a viable bacterial population capable of iron-reduction. This sandy clay layer, despite a pH value of 10.5, appears to be acting as a natural reactive zone beneath the waste as it is accumulating chromium. It is thought that the mechanism of Cr(VI) reduction is most likely to be an abiotic reaction with the Fe(II) present in the soil, and that Fe(II) in the soil is being replenished by microbial iron-reduction (although the rate of replenishment is unknown).

Keywords: Anaerobe; Alkaliphile; Bacteria; Chromate; Contaminated land; COPR; Iron-reduction; Microbial-reduction

Geeta S. Nagvenkar[1] and N. Ramaiah[1]. ([1]National Institute of Oceanography, Council of Scientific and Industrial Research (CSIR), Dona Paula, Goa, 403004, India). **Arsenite tolerance and biotransformation potential in estuarine bacteria. Ecotoxicology, Volume 19(4) (2010): 604-613**

Bacterial isolates from water and sediment samples from freshwater, estuarine and marine regions were tested for their growth in the presence of different concentrations of arsenic. Despite the generation times being longer in case of all bacterial isolates tested in nutrient broth with 200 ppm Arsenite (As^{3+}), many of them were able to attain log phase and substantial growth variously between 36 and 96 h. The isolates tolerating ≥200 ppm arsenic (As) were found to belong to Enterobacteriaceae, *Pseudomonas*, *Corynebaterium*, *Xanthomonas*, *Acinetobacter*, *Flavimonas* and *Micrococcus*. Some of these environmental strains tolerant to 1,000 ppm arsenic were tested to realize their potential to detoxify arsenic. The rate of As biotransformation was faster by many of these strains. The percent of arsenite biotransformed/removed from the growth medium was the highest by a strain of Enterobacteriaceae (as much as 92% of the As in the growth medium by 120 h) followed by that of *Corynebaterium* and *Acinetobacter* strains. From these observations it is clear that many environmental strains are capable of quite rapid biotransformation of As. Contamination of drinking water by toxic metalloid arsenic affects thousands of people worldwide. Many environmental isolates of bacteria which detoxify this metalloid would serve beneficial in the depuration processes. We suggest that only such strains capable of high tolerance to toxic arsenite, would biotransform As in polluted estuarine environments and would prove useful in As bioremediation applications.

Keywords: Arsenic tolerant bacteria - Toxic metalloid - Estuarine - As biotransformation - Arsenite - Enterobacteriaceae - *Coynebacterium* sp.

Xinde Cao[1, 3], Lena Ma[3], Aziz Shiralipour[2] and Willie Harris[3]. ([1]School of Environmental Science and Engineering, Shanghai Jiao Tong University, Shanghai, 200240, China, [2]Center for Natural Resources, University of Florida, Gainesville, FL 32611, USA, [3]Soil and Water Science Department, University of Florida, Gainesville, FL 32611, USA). **Biomass reduction and arsenic transformation during composting of arsenic-rich hyperaccumulator *Pteris vittata* L. Environmental Science and Pollution Research, Volume 17(3) (2010): 586-594**

Background, aim, and scope

Composting is being proposed as a pretreatment step before disposal of metal-rich biomass after phytoextraction process. This study determined the biomass reduction and arsenic transformation during composting As-rich biomass of hyperaccumulator Chinese brake fern (*Pteris vittata* L.).

Materials and methods

High-As fern biomass containing 4,600 mg As kg^{-1} was composted for 120 days in a laboratory-scale composter under aerated condition. Solid As speciation was determined using X-ray diffraction (XRD) and scanning electron microscopy equipped with X-ray energy dispersive elemental spectroscopy (SEM-EDS), while liquid As speciation was evaluated by high-performance liquid chromatography coupled with inductively coupled plasma mass spectrometry.

Results and discussion

Composting reduced the fern biomass by 38%, comparable to 35% reduction of the low-As fern biomass containing □12 mg As kg^{-1}, indicating that the high As concentration in *P. vittata* had little detrimental effect on microorganisms involved in composting. Total As on a per composter basis and water-soluble As in composted biomass were lowered by 25% and 32%, respectively. Arsenic loss from the biomass resulted mainly from generation of compost leachate, with less from the As volatilization; whereas As immobilization was due to oxidation of As(III) to As(V), followed by precipitation of hoernesite [$Mg_3(AsO_4)_2 \cdot 8H_2O$] which was evidenced by XRD and SEM-EDS analysis.

Conclusions and perspectives

Results from this study indicate that composting As-rich fern significantly reduced its biomass, As content, and water-soluble As. It seems that composting can be an effective step for treating As-rich biomass before disposal in landfill.

Keywords: Arsenic-rich biomass - Arsenic speciation - Composting - Phytoextraction - *Pteris vittata* L - Waste disposal

Itzamná Baqueiro-Peña[a], Gabriela Rodríguez-Serrano[a], Eduardo González-Zamora[b], Christopher Augur[c], Octavio Loera[a] and Gerardo Saucedo-Castañeda[a]. ([a]Department of Biotechnology, Autonomous Metropolitan University, Campus Iztapalapa, Av. San Rafael Atlixco 186, Col. Vicentina C.P. 09340, Iztapalapa, DF, Mexico, [b] Department of Chemistry, Autonomous Metropolitan University, Campus Iztapalapa, Av. San Rafael Atlixco 186, Col. Vicentina C.P. 09340, Iztapalapa, DF, Mexico, [c] Laboratoire IRD-IMEP, Université Paul Cézanne, Faculté des Sciences et Techniques de St. Jérôme, 13397 Marseille Cedex 20, France). **Biotransformation of ferulic acid to 4-vinylguaiacol by a wild and a diploid strain of *Aspergillus niger* . Bioresource Technology, Volume 101(12) (2010): Pages 4721-4724**

Ferulic acid biotransformation has a number of interesting industrial uses. Ferulic acid biotransformation by the wild strain *Aspergillus niger* C28B25 and a diploid strain DAR2, obtained by parasexual recombination, was studied. The wild strain of *A. niger* C28B25

biotransforms ferulic acid to vanillic acid (VA); while the diploid strain DAR2 preferentially decarboxylates ferulic acid to 4-vinylguaiacol (4VG). The latter was identified by mass spectroscopy, ^{1}H and ^{13}C nuclear magnetic resonance spectroscopy, and quantified by HPLC. The diploid strain *A. niger* DAR2 and the wild strain showed a ferulic acid conversion of 64% and 36%, respectively. Molar yields show that the formation of 4VG was preferred, being as much as 4.4 times higher than the formation of VA in diploid strain cultures. Differential regulation of enzymes involved in the biotransformation of ferulic acid may explain the accumulation of 4VG by diploid DAR2. This strain produced both 4VG and VA.

Keywords: *Aspergillus niger*; Ferulic acid; 4-Vinylguaiacol; Vanillic acid

Yijun Dai[a], Yinjuan Zhao[a, 1], Wenjian Zhang[a], Cigang Yu[a], Weiwei Ji[a], Wenping Xu[a], Jueping Ni[b] and Sheng Yuan[a]. ([a] Jiangsu Engineering and Technology Research Center for Industrialization of Microbial Resources, Jiangsu Key Lab for Biodiversity and Biotechnology, College of Life Science, Nanjing Normal University, Nanjing 210046, PR China, [b] Jiangsu Pesticide Research Institute, Nanjing 210036, PR China). **Biotransformation of thianicotinyl neonicotinoid insecticides: Diverse molecular substituents response to metabolism by bacterium *Stenotrophomonas maltophilia* CGMCC 1.1788. Bioresource Technology, Volume 101(11) (2010): 3838-3843**

The carbon atom that neighbors the tertiary amine attached to the 6-chloro-3-pyridinylmethyl moiety is the key active site in the hydroxylation of the neonicotinoids imidacloprid and thiacloprid as well as in the demethylation of acetamiprid by *Stenotrophomonas maltophilia* CGMCC 1.1788. In this study, thianicotinyl neonicotinoid insecticides having diverse molecular substituents were biotransformed by *S. maltophilia* CGMCC 1.1788. The results indicated that the substitution of 6-chloropyridyl in imidacloprid with 2-chlorothiazol in imidaclothiz did not affect the hydroxylation of imidaclothiz and its hydroxylated site, while the oxadiazinane ring in thiamethoxam was not hydroxylated or opened. Moreover, the *N*-methyl group in clothianidin and thiamethoxam was not demethylated by *S. maltophilia* CGMCC 1.1788. The biotransformation of imidaclothiz was inhibited by piperonyl butoxide, implying that both hydroxylation and dehydrogenation are mediated by a P450 monooxygenase. The bioassay results suggested that the activity of 5-hydroxy and olefin imidaclothiz was similar but less than that of imidaclothiz against the horsebean aphid *Aphis craccivora* and mosquito larva *Culex pipiens*, while 5-hydroxy IMT showed weak activity against the brown planthopper *Nilaparvata lugens*.

Keywords: *Stenotrophomonas maltophilia*; Neonicotinoid; Hydroxylation; Dehydrogenation; Bioefficacy

Mark L. Thompson[1], Ray Marriott[2], Adam Dowle[3] and Gideon Grogan[1]. ([1]York Structural Biology Laboratory, Department of Chemistry, University of York, YO10 5YW York, UK, [2]Department of Chemistry, University of York, YO10 5DD York, UK, [3]Technology Facility, Department of Biology, University of York, YO10 5YW York, UK). **Biotransformation of β-myrcene to geraniol by a strain of *Rhodococcus erythropolis* isolated by selective enrichment from hop plants. Applied Microbiology and Biotechnology, Volume 85(3) (2010): 721-730**

The biocatalytic generation of high-value chemicals from abundant, cheap and renewable feedstocks is an area of great contemporary interest. A strain of *Rhodococcus erythropolis*

designated MLT1 was isolated by selective enrichment from the soil surrounding hop plants, using the abundant triene β-myrcene from hops as a sole carbon source for growth. Resting cells of the organism were challenged with β-myrcene, and the major product of biotransformation was determined by mass spectrometric analysis to be the monoterpene alcohol geraniol. Controls demonstrated that the product was biogenic and that an aerobic environment was required. The ability to transform β-myrcene was shown to be restricted to cells that had been grown on this substrate as sole carbon source. Pre-incubation of cells with the cytochrome P450 inhibitors metyrapone or 1-aminobenzotriazole reduced geraniol production by 23% and 73% respectively, but reduction in activity was found not to correlate with the inhibitor concentration. A comparative analysis of insoluble and soluble cell extracts derived from cells of MLT1 grown on either β-myrcene or glucose revealed at least four proteins that were clearly overproduced in response to growth on β-myrcene. Mass spectrometric analysis of tryptic digests of three of these protein bands suggested their identities as an aldehyde dehydrogenase, an acyl-CoA dehydrogenase and a chaperone-like protein, each of which has a precedented role in hydrocarbon metabolism clusters in *Rhodococcus* sp. and which may therefore participate in a β-myrcene degradation pathway in this organism.

Keywords: Hops - Enrichment - *Rhodococcus* - Biotransformation - Myrcene – Geraniol

Hongjuan Li[1], Xuemei Li[2], Yanqing Duan[3], Ke-Qin Zhang[1] and Jinkui Yang[1]. ([1]Laboratory for Conservation and Utilization of Bio-Resources, and Key Laboratory for Microbial Resources of the Ministry of Education, Yunnan University, Kunming, 650091, People's Republic of China, [2]Yunnan Academy of Tobacco Science, Kunming, 650106, People's Republic of China, [3]Technology Centre of Hongyun Honghe Tobacco (Group) Co, Ltd, Kunming, 650202, People's Republic of China). Biotransformation of nicotine by microorganism: the case of *Pseudomonas* spp. Applied Microbiology and Biotechnology, Volume 86(1) (2010): 11-17

Several bacterial species are capable of using nicotine, the main alkaloid in tobacco plants, as a substrate for growth. The dominant species include members of two genera, *Pseudomonas* and *Arthrobacter*. The degradation pathway and genetic structure of nicotine catabolism in *Arthrobacter nicotinovorans* were recently reviewed (Brandsch Appl Microbiol Biotechnol 69:493–498, 2006). Here, we present up-to-date information on biodegradation of nicotine by *Pseudomonas* spp. Species in this genus capable of degrading nicotine are summarized and analyzed phylogenetically. Their metabolic intermediates and nicotine degradation-related genes were summarized, and the nicotine-biotransformation pathways were compared and discussed.

Keywords: Nicotine degradation - *Pseudomonas* spp. - Metabolic products - Nicotine degradation-related genes - Biotransformation pathway

Jessica Amadio[1] and Cormac D. Murphy[1]. ([1]School of Biomolecular and Biomedical Science, Centre for Synthesis and Chemical Biology, Ardmore House, University College Dublin, Dublin 4, Ireland). **Biotransformation of fluorobiphenyl by *Cunninghamella elegans*. Applied Microbiology and Biotechnology, Volume 86(1) (2010): 345-351**

The fungus *Cunninghamella elegans* is a useful model of human catabolism of xenobiotics. In this paper, the biotransformation of fluorinated biphenyls by *C. elegans* was investigated by analysis of the culture supernatants with a variety of analytical techniques. 4-Fluorobiphenyl was

principally transformed to 4-fluoro-4′-hydroxybiphenyl, but other mono- and dihydroxylated compounds were detected in organic extracts by gas chromatography–mass spectrometry. Additionally, fluorinated water-soluble products were detected by ^{19}F NMR and were identified as sulphate and β-glucuronide conjugates. Other fluorobiphenyls (2-fluoro-, 4,4′-difluoro- and 2,3,4,5,6-pentafluoro-biphenyl) were catabolised by *C. elegans*, yielding mono- and dihydroxylated products, but phase II metabolites were detected from 4,4′-difluorobiphenyl only.

Keywords: Fluorine - Biphenyl - Metabolism - F-19 NMR

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Cell-free synthetic (enzymatic) pathway biotransformation (SyPaB) is the assembly of a number of purified enzymes (usually more than 10) and coenzymes for the production of desired products through complicated biochemical reaction networks that a single enzyme cannot do. Cell-free SyPaB, as compared to microbial fermentation, has several distinctive advantages, such as high product yield, great engineering flexibility, high product titer, and fast reaction rate. Biocommodities (e.g., ethanol, hydrogen, and butanol) are low-value products where costs of feedstock carbohydrates often account for ~30-70% of the prices of the products. Therefore, yield of biocommodities is the most important cost factor, and the lowest yields of profitable biofuels are estimated to be ca. 70% of the theoretical yields of sugar-to-biofuels based on sugar prices of ca. US$ 0.18 per kg. The opinion that SyPaB is too costly for producing low-value biocommodities are mainly attributed to the lack of stable standardized building blocks (e.g., enzymes or their complexes), costly labile coenzymes, and replenishment of enzymes and coenzymes. In this perspective, I propose design principles for SyPaB, present several SyPaB examples for generating hydrogen, alcohols, and electricity, and analyze the advantages and limitations of SyPaB. The economical analyses clearly suggest that developments in stable enzymes or their complexes as standardized parts, efficient coenzyme recycling, and use of low-cost and more stable biomimetic coenzyme analogs, would result in much lower production costs than do microbial fermentations because the stabilized enzymes have more than 3 orders of magnitude higher weight-based total turn-over numbers than microbial biocatalysts, although extra costs for enzyme purification and stabilization are spent.

Keywords: biocatalyst • bioelectricity • biocommodity • cell-free synthetic biology • synthetic pathway biotransformation (SyPaB) • weight-based total turn-over number (TTN_W)

W. L. Ma[1], C. Y. Yan[1, 2], J. H. Zhu[1], G. Y. Duan[1] and R. M. Yu[1]. ([1]College of Pharmacy, Jinan University, Guangzhou, 510632, China, [2]College of Pharmacy, Guangdong Pharmaceutical University, Guangzhou, 510006, China). Biotransformation of Paeonol and Emodin by Transgenic Crown Galls of *Panax quinquefolium*. Applied Biochemistry and Biotechnology, Volume 160(5) (2010): 1301-1308

Two aromatic substrates, paeonol (**1**) and emodin (**2**), were biotransformed by using transgenic crown galls of *Panax quinquefolium*. Four biotransformed products (**3**–**6**) were isolated and identified by physicochemical and spectral methods. A β-glucoside (**3**, 73.2% of biotransformation yield) and a 1-(2,4-dimethoxyphenyl)- ethanone (**4**, 8.03%) were isolated from the suspension cultures after 7-day incubation of substrate **1**. Upon administration of substrate **2**, another β-glucoside [emodin-6-O-β-D-glucopyranoside (**5**), 19.2%] and a hydroxylated derivative, citreorosein (**6**, 54.6%), were also obtained. The results demonstrate that transgenic crown galls of *P. quinquefolium* have the capacities to catalyze glycosylation, hydroxylation, and methylation reactions in the plant cells on those aromatic compounds.

Keywords: Transgenic crown galls - *Panax quinquefolium* - Biotransformation - Paeonol - Emodin

Biomarker

Kimberly S. Beltran1, [2] and Glorina N. Pocsidio[1]. ([1]Institute of Biology, College of Science, University of the Philippines, Diliman, Quezon City, 1101, Philippines, [2]Department of Biology, College of Arts and Sciences, University of the Philippines, Ermita, Manila, Philippines). Acetylcholinesterase activity in Corbicula fluminea Mull., as a biomarker of organophosphate pesticide pollution in Pinacanauan River, Philippines. Environmental Monitoring and Assessment, Volume 165(1-4) (2010): 331-340

Organophosphates are known to inhibit the enzyme acetylcholinesterase. In this study, the AChE activity from the total soft tissues of *Corbicula fluminea* Mull. was used as a biomarker of organophosphate pollution in Pinacanauan River. Clams were collected from two different sites and at different seasons of the year. A colorimetric assay on the total soft tissues of the clams showed a directly proportional relationship between enzyme activity and condition of the riverine system. In vitro experiments on the total soft tissue, adductor muscles, digestive glands, and gills were conducted to assess the degree of localization of AChE as well as the sensitivity and tolerance of the enzymes in these tissues to varying concentrations of malathion. The degree of enzyme localization from highest to lowest is as follows: adductor muscle > gills > digestive gland whereas sensitivity to OP from greatest to least is: gills > adductor muscles > digestive gland.

Keywords: Acetylcholinesterase - *Corbicula fluminea* - Malathion - Pinacanauan River

Andrea Luna-Acosta[1], Paco Bustamante[1], Joachim Godefroy[1], Ingrid Fruitier-Arnaudin[1] and HÃ©lÃ¨ne Thomas-Guyon[1]. ([1] Littoral, Environnement et SociÃ©tÃ©s (LIENSs), UMR 6250, UniversitÃ© de La Rochelle—CNRS, 2, Rue Olympe de Gouges, 17000 La Rochelle, France). Seasonal variation of pollution biomarkers to assess the impact on the health status of juvenile Pacific oysters Crassostrea gigas exposed in situ. Environmental Science and Pollution Research, Volume 17(4) (2010): 999-1008

Background, aim, and scope

In this study, a suite of sublethal stress biomarkers were analyzed in juveniles of the sentinel species, the Pacific oyster *Crassostrea gigas*, with a view to using them as pollution monitoring tools. The aim of this work was (1) to study baseline seasonal variations of biomarkers in different body compartments of *C. gigas* in the reference site and, after selecting biomarkers presenting no seasonal variations, (2) to compare responses of these biomarkers between contaminated and reference sites.

Materials and methods

Juvenile oysters were transplanted from Bouin (France), a reference site, to three different sites in Marennes-Oleron Bay (France), located in another water body and next to different contamination sources. Animals were exposed in situ for 3 months in summer, autumn, and winter. The following biomarkers were measured: superoxide dismutase (SOD) and glutathione peroxidase (GPx) in gills and digestive gland and lysozyme and phenoloxidase (PO) in plasma.

Results

No significant seasonal variations for SOD in gills and digestive gland, GPx in gills, and PO in plasma were observed in the reference site. Significant differences in enzyme activity were observed between contaminated and reference sites for SOD in gills and digestive gland and PO in plasma, depending on the body compartment, the season, and/or the site.

Conclusions

In conclusion, these data suggest the potential application of these biomarkers in *C. gigas* to provide ecologically relevant information and, therefore, to be used as biomarkers in coastal pollution monitoring.

Keywords: Biomarkers - Pollution - Monitoring - Antioxidant enzymes - Immune defenses - *Crassostrea gigas*

Anita Jemec[1], Damjana Drobne[2], Tatjana Tišler[1] and Kristina Sepčić[2]. ([1]National Institute of Chemistry, Hajdrihova 19, 1000 Ljubljana, Slovenia, [2]Department of Biology, Biotechnical Faculty, University of Ljubljana, Večna pot 111, 1000 Ljubljana, Slovenia). **Biochemical biomarkers in environmental studiesâ€"lessons learnt from enzymes catalase, glutathione S-transferase and cholinesterase in two crustacean species. Environmental Science and Pollution Research, Volume 17(3) (2010): 571-581**

Background, aim and scope

For reliable environmental risk assessment of pollutants, knowledge on the effects at different levels of biological organisation is needed. During the early days of biomarker research in environmental studies approximately two decades ago, biochemical biomarkers were considered as the most promising tool for such purposes. Among these, three enzymes have often been studied: catalase (CAT), glutathione *S*-transferase (GST) and cholinesterase (ChE). However, despite their intensive research, their measurements in invertebrates have not been commonly applied in environmental risk assessment (ERA) or for regulatory purposes.

Main features

In the present review, we summarise our past experiences in biochemical biomarker research in two crustacean species: water flea *Daphnia magna* and terrestrial isopod *Porcellio scaber*. This is to orientate their use and to provide recommendations for the use of novel biomarkers in environmental studies, such as proteomic or genomic responses.

Results and discussion

We assessed the intrinsic properties of biochemical biomarkers CAT, GST and ChE in the *D. magna* and the isopod *P. scaber*. It was found that they are not in agreement with the expectations that were previously given for their use in environmental studies. To advance their use in environmental risk assessment, we suggest that based on their properties, their role should be more specifically defined. ERA includes several distinct steps, among them hazard identification, effect assessment and finally risk characterisation, each of which requires a different type of toxicity data. We recommend that the use of biochemical markers is most appropriate for hazard identification because this is a procedure whose purpose is to characterise the potential hazard of the substance in question and is more flexible in terms of using different tools. Furthermore, our results imply that biochemical markers are not always more sensitive than whole-organism responses, as was anticipated. Their sensitivity depends on the mode of action, duration of exposure and test species. Therefore, we suggest that combining both a battery of biomarkers from different levels of biological organisation and an array of biomarkers within a single level could identify hazard adequately.

Conclusions

The lesson learnt from biochemical biomarkers in environmental studies utilizing crustacean model species is that, for successful application of each group of biomarkers, their intrinsic properties are needed to be known before an (eco)toxicity study is designed. We suggest that a substantial body of experience obtained with biochemical biomarkers should be exploited to new emerging biomarkers in environmental studies in order to facilitate their application.

Recommendations and perspectives

The future of biomarkers lies in a combination of traditional biochemical and new-generation biomarkers. The latter are not only a potential replacement for existing biomarkers but will also provide new knowledge which might encourage renewed research and development of traditional biomarkers. For research purposes, complete ecotoxicity information should include contributions from molecular fingerprint of an organism, as well as whole organism, population and ecosystem responses. Still, the type of biomarkers used for routine purposes will depend on their reproducibility, their ease of use, robustness, affordability of the methodology and the type of chemicals, organisms and ecosystem of interest.

Keywords: Biochemical biomarker - Catalase - Cholinesterase - *Daphnia magna* - Environmental risk assessment - Glutathione *S*-transferase - Hazard - Marker - Metals - Pesticides - Terrestrial invertebrate *Porcellio scaber* - Toxicogenomics - Toxicoproteomics

Biofertilizer

Shuhe Wei[a], Yunmeng Li[a, b], Qixing Zhou[a], Mrittunjai Srivastava[c], Siuwai Chiu[d], Jie Zhan[e], Zhijie Wu[a] and Tieheng Sun[a]. ([a] Key Laboratory of Terrestrial Ecological Process, Institute of Applied Ecology, Chinese Academy of Sciences, Shenyang 110016, PR China, [b] Graduate School of Chinese Academy of Sciences, Beijing 100039, PR China, [c] North Florida Research and Education Center, University of Florida, Quincy, FL 32351-5677, USA, [d] Department of Biology, The Chinese University of Hong Kong, Shatin, N.T., Hong Kong SAR, PR China, [e] Department of Biotechnology, Liaoning University of Traditional Chinese Medicine, Shenyang 110101, PR China). Effect of fertilizer amendments on phytoremediation of Cd-contaminated soil by a newly discovered hyperaccumulator *Solanum nigrum* L. Journal of Hazardous Materials, Volume 176(1-3) (2010): 269-273

Phytoremediation is a cost-effective, simple and sustainable beneficiary technique to purify the polluted environment. *Solanum nigrum* L., a newly found cadmium (Cd) hyperaccumulator, has shown the potential to remediate Cd-contaminated soils. Present study investigated the effects of fertilizer amendments on the Cd uptake by *S. nigrum*. Chicken manure and urea are usual agricultural fertilizers and more environmental friendly. The results showed that Cd concentrations in shoots of *S. nigrum* were significantly decreased ($p < 0.05$) by 28.2–34.6%, as compared to that of without the addition of chicken manure, but not the case for urea treatment. However, Cd extraction capacities (μg pot^{-1}) in shoot biomass of *S. nigrum* were significantly increased ($p < 0.05$) due to increased shoot biomass. In addition, available Cd concentration in soil significantly decreased due to addition of chicken manure. Thus, urea might be a better fertilizer for strengthening phytoextraction rate of *S. nigrum* to Cd, and chicken manure may be a better fertilizer for phytostabilization.

Keywords: Cadmium; Fertilizer; Phytoextraction; Phytostability; *Solanum nigrum*

P.R. Warman[a] and M.J. AngLopez[a]. ([a] Nova Scotia Agricultural College, P.O. Box 550, Truro, N.S., Canada B2N 5E3). Vermicompost derived from different feedstocks as a plant growth medium. Bioresource Technology, Volume 101(12) (2010): 4479-4483

This study determined feedstock effects on earthworm populations and the quality of resulting vermicomposts produced from different types of feedstocks using different vermicomposting durations. Feedstock combinations (Kitchen Paper Waste (KPW), Kitchen Yard Waste (KYW), Cattle Manure Yard Waste (CMY)), three durations of vermicomposting (45, 68 or 90 days), and two seed germination methods (with two concentrations of vermicompost) for radish, marigold and upland cress, served as the independent variables.

The worms (*Eisenia fetida*) doubled their weight by day 68 in KPW and CMY vermicomposts and day 90 KPW vermicompost produced the greatest weight of worms. The direct seed germination method (seeding into soil or vermicompost–soil mixtures) indicated that KPW and KYW feedstocks decreased germination compared to the control, even in mature vermicompost. Seed germination was greater in the water extract method; however, most of the vermicompost extracts suppressed germination of the three seed species compared to the water controls. Vermicomposts from all three feedstocks increased leaf area and biomass compared to the control, especially in the 10% vermicompost:soil mix. Thus, seed germination and leaf area or plant biomass for these three species are contrasting vermicompost quality indicators.

Keywords: Feedstock; Germination; Plant bioassay; Vermicompost maturity

Biocomposting

Ghulam Sarwar , Nazir Hussain , Fakhar Mujeeb , H. Schmeisky and Ghulam Hassan. Biocompost Application for the Improvement of Soil Characteristics and Dry Matter Yield of *Lolium perenne* (Grass). Asian Journal of Plant Sciences, Volume 2(2) (2003): 237-241

A pot experiment was conducted at University of Kassel, Witzenhausen Germany. Three types of soils were used for this purpose. Biocompost is the form of compost prepared from kitchen wastes. This compost was analyzed before application (pH = 7.7 and C: N ratio = 14.2:1). Biocompost was applied to each soil type at 5 and 10% of soil volume. *Lolium perenne* grass was grown in all the pots. The experiment had four replications. This grass was harvested at the age of one month and was oven dried at 60 ^{o}C. This plant material was then analyzed for organic matter; N, Ca, Mg, K and P contents. Soil samples were also taken from all the pots and analyzed for pH, EC, organic matter, N, C, C/N ratio and mineral nutrients (Ca, Mg, K, P and Cl). All data were analyzed statistically. It was observed that use of biocompost at both the levels (5 and 10%) enhanced the soil pH, EC, organic matter, N and C% with a net decrease in C/N ratio. Organic matter and N percentages in plant material were increased with the application of biocompost. Contents of mineral nutrients in the soil were also increased by the use/addition of biocompost. The resultant improvements in soils, contributed towards significant enhancement in dry matter yield of *Lolium perenne* grass.

R. Schulz[12] , H. Al-Najar[12], J. Breuer[13] and V. Römheld[12]. ([12]Institute of Plant Nutrition (330), University of Hohenheim, 70593 Stuttgart, Germany, [13]Institute of Agricultural Chemistry (710), University of Hohenheim, 70593 Stuttgart, Germany). The effect of bio-compost application on crop yield and nitrogen dynamic in the soil. Plant Nutrition, Volume 92 : 986-987

Since 1997 bio-compost with and without optimised mineral N fertilisation is applied to maize, winter wheat and summer barley (crop rotation) in a randomised field experiment on a Luvisol in south-west Germany. The aim of this experiment is to evaluate the effect of bio-compost application on crop yield and N-net mineralisation in soil. In the bio-compost application treatment (100 kg total **N ha^{-1} a^{-1}**, that is about 7.5 t DM) with optimised mineral N fertilisation yield of summer barley (1999) and maize (2000) was higher than in the optimised mineral N treatment without bio-compost application. Similar results were obtained with the higher bio-compost application rate (400 kg total **N ha^{-1} a^{-1}**, that is about 30 t DM) without additional mineral N fertilisation. The yield increase can be attributed to an improvement of soil structure in the Luvisol. During the vegetation period of summer barley net-N-mineralisation on the field plots of the bio-compost treatments with additional mineral N fertilisation was lower than in plots without additional mineral N application. The higher Nmin values after harvest (Sept. 1999) in bio-compost treatments with additional mineral N-fertilisation can be partially attributed to the remineralisation of immobilised N at the end of the growth period.

Key words: bio-compost - nitrogen mineralisation - nitrogen immobilisation - nitrogen dynamic

Biopesticides

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Insect pests persist in a wide-variety of agricultural, arboreal and urban environments. Effective control with fungal entomopathogens using inundation biocontrol requires an understanding of the ecology of the target insect, fungal pathogen, and the insect-pathogen interaction. Historically, the development of production and formulation processes for biocontrol fungi has primarily focused on reducing costs by maximizing the yield of infective propagules, increasing storage stability, and improving product form for ease of application. These goals are critical for commercialization but are often in conflict with environmental and ecological considerations. Critical parameters for selecting a fungal pathogen for use in inundation biocontrol include the cost-effective production of a stable, infective propagule that is suited for use in the environment where the insect must be controlled. Production processes can be manipulated nutritionally and environmentally to produce efficacious propagules or to direct fungal differentiation to propagule forms that may be better suited for use in specific environments. Formulation development must also consider ecological and environmental factors to maximize biocontrol efficacy. A basic understanding of the surface chemistries of the fungal propagule and insect, the interactions between a fungal propagule and the insect cuticle that lead to infection, and the impact of the environment on this interaction can aid in the development of effective formulations.

Keywords: Biocontrol - Fungi - Fermentation - Formulation - Conidia - Blastospores - Sclerotia - Mycoinsecticides

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Fungal entomopathogens have been used more frequently than other types of pathogens for classical biological control. Among 136 programs using different groups of arthropod pathogens, 49.3% have introduced fungal pathogens (including both the traditional fungi and microsporidia). The most commonly introduced species was *Metarhizium anisopliae* (Metschnikoff) Sorokin, with 13 introductions, followed by *Entomophaga maimaiga* Humber, Shimazu & Soper, which was released seven times. The majority of introduction programs have focused on controlling invasive species of insects or mites (70.7%) rather than on native hosts (29.4%). Almost half of the introductions of traditional fungi targeted species of Hemiptera and 75% of the microsporidia introduced have been introduced against lepidopteran species. The United States was the country where most introductions of fungi took place ($n = 24$). From 1993 to 2007, no arthropod pathogens were released in the US due to the rigorous regulatory structure, but in 2008 two species of microsporidia were introduced against the gypsy moth, *Lymantria*

dispar (L.). Establishment of entomopathogenic fungi in programs introducing traditional fungi was 32.1% and establishment was 50.0% for programs introducing microsporidia. In some programs, releases have resulted in permanent successful establishment with no non-target effects. In summary, classical biological control using fungal entomopathogens can provide a successful and environmentally friendly avenue for controlling arthropod pests, including the increasing numbers of invasive non-native species.

Keywords: Biological control - Microbial control - Entomopathogens - Fungi - Microsporidia

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Conservation biological control relies on modification of the environment or management practices to protect and encourage natural enemies that are already present within the system, thereby enhancing and improving their ability to control pest populations in a reliable way. Such strategies are only possible when based on a strong understanding of the ecology of the species concerned at the individual, community and landscape scale. Conservation biological control with entomopathogenic fungi includes the manipulation of both the crop environment and also habitats outside the crop. Further investment in conservation biological control with entomopathogenic fungi could make a substantial contribution to sustainable crop production either as stand alone strategies or, more importantly, in support of other biological and integrated pest management strategies.

Keywords: Ecology - Epizootiology - Entomophthorales - Hypocreales - Pest control - Conservation

Ali Dehestani[1, 2, 3], Kamal Kazemitabar[1], Gholamreza Ahmadian[2], Nadali Babaeian Jelodar[1], Ali Hatef Salmanian[3], Mehdi Seyedi[3], Heshmat Rahimian[4] and Seyedhadi Ghasemi[1, 2]. ([1]Department of Plant Breeding, Faculty of Agronomic Sciences, Sari Agricultural Sciences and Natural Resources University, P. O. Box 578, Sari, Iran, [2]Department of Molecular Genetics, National Institute for Genetic Engineering and Biotechnology (NIGEB), P. O. Box 14155-6343, Tehran, Iran, [3]Department of Plant Biotechnology, National Institute for Genetic Engineering and Biotechnology (NIGEB), P. O. Box 14155-6343, Tehran, Iran, [4]Department of Plant Protection, Faculty of Agronomic Sciences, Sari Agricultural Sciences and Natural Resources University, P. O. Box 578, Sari, Iran). **Chitinolytic and antifungal activity of a *Bacillus pumilus* chitinase expressed in *Arabidopsis.* Biotechnology Letters, Volume 32(4) (2010): 539-546**

The *Bacillus pumilus* SG2 chitinase gene (*ChiS*) and its truncated form lacking chitin binding (ChBD) and fibronectin type III (FnIII) domains were transformed to *Arabidopsis* plants and the expression, functionality and antifungal activity of the recombinant proteins were investigated. Results showed that while the two enzyme forms showed almost equal hydrolytic activity toward colloidal chitin, they exhibited a significant difference in antifungal activity. Recombinant ChiS in plant protein extracts displayed a high inhibitory effect on spore germination and radial

growth of hyphae in *Alternaria brassicicola*, *Fusarium graminearum* and *Botrytis cinerea*, while the activity of the truncated enzyme was strongly abolished. These findings demonstrate that ChBD and FnIII domains are not necessary for hydrolysis of colloidal chitin but play an important role in hydrolysis of chitin–glucan complex of fungal cell walls. Twenty microgram aliquots of protein extracts from ChiS transgenic lines displayed strong antifungal activity causing up to 80% decrease in fungal spore germination. This is the first report of a *Bacillus pumilus* chitinase expressed in plant system.

Keywords: Antifungal activity - *Arabidopsis thaliana* - *Bacillus pumilus* - Chitin binding domain - Chitinase activity

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The biocontrol bacterium *Lysobacter* sp. SB-K88 suppresses damping-off disease in sugar beet and spinach caused by *Aphanomyces cochlioides* and *Pythium* sp. through characteristic plant colonization and antibiosis against the pathogens. This study aimed to unravel further details on mode of antagonism of SB-K88 against a damping-off pathogen *A. cochlioides* AC-5. The SB-K88 substantially inhibited growth and decomposed AC-5 mycelia and suppressed the release of zoospores from the hyphae. The excised root tips of sugar beet seedlings from seeds previously inoculated with SB-K88 were less attractive to AC-5 zoospores. Although aerial growth was not affected, however, root hairs of SB-K88 inoculated sugar beet seedlings were remarkably shorter and thicker than those of uninoculated control. When exposed to zoospores, the SB-K88 inhibited motility of zoospores and/or caused lysis, and then aggregated around the dead cystospores or lysed residues within 3–6 h likely to be micro-predatory behavior to a eukaryotic organism. Confocal laser scanning microscopic analysis revealed that number of lipid bodies and activities of mitochondria were markedly increased in the affected hyphae compared with control hyphae as visualized by established vital stains. Taken together, these results suggest that *Lysobacter* sp. SB-K88 suppresses damping-off diseases through exerting multifaceted antagonistic effects against the peronosporomycetes.

Keywords: *Lysobacter* sp. - Chemotaxis - *Aphanomyces cochlioides* - Mitochondrial activity - Lipid bodies – Micropredation

Teresa Manso[1], Carla Nunes[1], Sara Raposo[1] and Maria Emília Lima-Costa[1]. ([1]CDCTPV-Food Safety and Technology Research Center, Universidade do Algarve, Campus de Gambelas, 8005-139 Faro, Portugal). **Production of the biocontrol agent *Pantoea agglomerans* PBC-1 in a stirred tank reactor by batch and fed-batch cultures. World Journal of Microbiology and Biotechnology, Volume 26(4) (2010): 725-735**

Concerns about food safety as well as the development of resistance to many fungicides by major postharvest pathogens have increased recently. Biological control, using microorganisms antagonistic to the fungal plant pathogens, appears to be promising as an alternative to fungicides. The microbial biocontrol agent has to be produced on an industrial scale, maintaining its biocontrol efficacy. The purpose of the current study was to optimize the conditions for

microbial biomass production of the biocontrol agent *Pantoea agglomerans* PBC-1 in a 2-l mechanically stirred reactor (STR), defining mixing and mass transfer technological parameters and the growth kinetics for different saccharides. In the batch mode, different impellers and spargers were tested. Despite the oxygen mass transfer improvement achieved with marine propeller combined with porous sparger, the biomass did not increase, if compared with the use of a Rushton turbine and L-sparger, pointing out the relevance of a radial flux for better broth homogenization. Different carbon sources were used: sucrose, glucose and fructose; each of which led to viable populations 3.9×10^9, 1.4×10^9, 3.9×10^9 c.f.u/ml, respectively, after 20 h of incubation. Fed-batch technology allows the maintenance of high cell viability for longer periods of time in the stationary growth phase, which can be crucial for the scale-up of biocontrol agent production process that is achieved together with a reduction of 85% on the incidence caused by the pathogens, brought about by fresh microbial biomass preparation on artificially wounded apples or oranges, stored for 7 days at 25°C against *Penicillium expansum* and *Penicillium digitatum.*

Keywords : Batch culture - Fed-batch culture - Mixing - Mass transfer - Cell growth - Stirred bioreactor - Biocontrol agent - *Pantoea agglomerans*

Soma Roy[1], Kiranmayee Rao[2], Ch. Bhuvaneswari[2], Archana Giri[2] and Lakshmi Narasu Mangamoori[2]. ([1]Biotechnology Department, Chaitanya Bharathi Institute of Technology, Hyderabad, India, [2]Center for Biotechnology, Institute of Science and Technology, Jawaharlal Nehru Technological University, Hyderabad, Andhra Pradesh, 500 085, India). **Phytochemical analysis of *Andrographis paniculata* extract and its antimicrobial activity. World Journal of Microbiology and Biotechnology, Volume 26(1) (2010): 85-91**

The present study describes the phytochemical profile and antimicrobial activity of *Andrographis paniculata*. For the present investigation, two samples of *A. paniculata* extracts, obtained by extraction in chloroform and chloroform + HCl, respectively, were compared for their antimicrobial activity and further subjected to GC-MS analysis to find out the nature of the compounds responsible for the antimicrobial activity. The antibacterial activities were assessed by measuring the diameter of the inhibition zones, MIC and MBC values. Compared to the chloroform + HCl extract, the chloroform extract showed better antimicrobial activity against all the nine pathogenic bacterial strains tested. The chloroform extract was observed to be active against the opportunistic and pathogenic gram-negative bacteria, indicating its potential application related to noscomial infections. GC-MS results revealed phenols, aromatic carboxylic acids and esters in the chloroform extract to be the molecules responsible for the antimicrobial activity of *A. paniculata*. This is the first report on analysis of antimicrobial components from *A. paniculata*, and our results confer the utility of this plant extract in developing a novel broad spectrum antimicrobial agent.

Keywords : *Andrographis paniculata* - Antimicrobial activity - Gram-positive bacteria - Gram-negative bacterial - GC-MS

Steven D. Frank[a]. ([a]Department of Entomology, North Carolina State University, Campus Box 7613, Raleigh, NC 27695-7613, USA). **Biological control of arthropod pests using**

banker plant systems: Past progress and future directions. Biological Control, Volume 52(1) (2010): 8-16

The goal of banker plant systems is to sustain a reproducing population of natural enemies within a crop that will provide long-term pest suppression. The most common banker plant system consists of cereal plants infested with *Rhopalosiphum padi* L. as a host for the parasitoid *Aphidius colemani* L. *Aphidius colemani* continually reproduce and emerge from the banker plants to suppress aphid pests such as *Aphis gossypii* Glover and *Myzus persicae* Sulzer. Banker plant systems have been investigated to support 19 natural enemy species targeting 11 pest species. Research has been conducted in the greenhouse and field on ornamental and food crops. Despite this there is little consensus of an optimal banker plant system for even the most frequently targeted pests. Optimizing banker plant systems requires future research on how banker plants, crop species, and alternative hosts interact to affect natural enemy preference, dispersal, and abundance. In addition, research on the logistics of creating, maintaining, and implementing banker plant systems is essential. An advantage of banker plant systems over augmentative biological control is preventative control without repeated, expensive releases of natural enemies. Further, banker plants conserve a particular natural enemy or potentially the 'right diversity' of natural enemies with specific alternative resources. This may be an advantage compared to conserving natural enemy diversity per se with other conservation biological control tactics. Demonstrated grower interest in banker plant systems provides an opportunity for researchers to improve biological control efficacy, economics, and implementation to reduce pesticide use and its associated risks.

Keywords: Alternative prey; *Aphidius colemani*; *Aphidoletes aphidimyza*; *Aphis gossypii*; Augmentative biological control; *Bemisia tabaci*; Conservation biological control; *Encarsia formosa*; Greenhouse; Integrated pest management; *Myzus persicae*; Open-rearing system; Release strategy

Leigh J. Pilkington[a], Gerben Messelink[b], Joop C. van Lenteren[c] and Kristian Le Mottee[d]. ([a] EH Graham Centre for Agricultural Innovation, Gosford Primary Industries Institute, Locked Bag 26, Gosford, NSW 2250, Australia, [b] Wageningen UR Greenhouse Horticulture, P.O. Box 20, 2265 ZG Bleiswijk, The Netherlands, [c] Laboratory of Entomology, Wageningen University, P.O. Box 8031, 6700 EH, Wageningen, The Netherlands, [d] NSW Department of Primary Industries, Gosford Primary Industries Institute, Locked Bag 26, Gosford, NSW 2250, Australia). **"Protected Biological Control" – Biological pest management in the greenhouse industry. Biological Control, Volume 52(3) (2010): 216-220**

This paper briefly describes the foundations and characteristics of biological control in protected cropping and what drivers are behind adoption of this management system within this industry. Examining a brief history of biological control in greenhouses and what makes it a successful management strategy within the industry, the authors describe the rapid growth of biological control in parts of Europe and what this may mean for the industry in other parts of the world. The reaction of the greenhouse industry to several consumer led campaigns aimed at reducing the incidence of pesticides in the marketplace may be replicated in many other parts of the world. The size and robustness of the biological control industry in greenhouses, which is a reflection of the inherent characteristics of this industry that lends itself to biological control, is strong and growing with indications that this trend will be followed in many areas of the world.

Keywords: Greenhouse; Protected agriculture; Glasshouse; Natural enemies; Integrated pest management

Oktay Erdogan[a] and Kemal Benlioglu[b], ([a] Cotton Research Institute, Nazilli, Aydin, Turkey, [b] Adnan Menderes University, Faculty of Agriculture, Plant Protection Dept., 09100 Aydin, Turkey). **Biological control of Verticillium wilt on cotton by the use of fluorescent *Pseudomonas* spp. under field conditions. Biological Control, Volume 53(1) (2010): 39-45**

Four out of 59 fluorescent *Pseudomonas* spp. strains collected from cotton and weed rhizosphere were selected based on the following criteria: (1) inhibition of *Verticillium dahliae in vitro*, (2) disease suppression on two cotton cultivars grown from bacterized seeds using stem-injection with the conidia of *V. dahliae*, and (3) seedling vigor test (dry weight) under greenhouse conditions. Four selected *Pseudomonas* strains isolated from *Xanthium strumarium* (FP22), *Portulaca* sp. (FP23), *Gossypium hirsitum* (FP30), and *Convolvulus arvensis* (FP35), as well as the known biocontrol agent *Serratia plymuthica* (HRO-C48), were further tested for the impact on Verticillium wilt, growth parameters of cotton, and yield in a naturally infested field. The reduction of AUDPC by the seed bacterization with FP22, FP23, FP30, FP35, and HRO-C48 compared to non-bacterized control ranged from 39.2% to 50.9% and 22.1% to 36.8% in trials done in 2005 and 2006, respectively. The growth parameters (plant height, number of nodes on main stem, and NAWF-nodes above white flower) were significantly higher in seed bacterized plants compared to the untreated control. In the 2005 field trial, the increase of seed cotton yield by the treatment with four *Pseudomonas* strains and HRO-C48 ranged from 13.1% to 22.3% in Sayar 314 and 4.2% to 12.8% in Acala Maxxa. Seed cotton yield was not significantly influenced by the 2006 treatments. Our results indicate that seed treatment of cotton plants with our *Pseudomonas* spp. strains and the known strain *Serratia plymuthica* can help in the biocontrol of *V. dahliae* and improve growth parameters in cotton fields.

Keywords: *Verticillium*; Cotton; Fluorescent *Pseudomonas*; *Serratia plymuthica*; Weed; Field trial

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Acacia cyclops is one of the 13 species of Australian acacias (Mimosaceae) that have become naturalized in South Africa and are now declared invasive weeds. *Dasineura dielsi*, a multivoltine midge that induces galls on the ovary of its host, has been introduced for biological control of *A. cyclops* in South Africa where it now colonizes 100% of the mature trees and substantially reduces their reproductive capabilities. Although *A. cyclops* is the primary host, galls are sometimes induced by on several other Australian *Acacia* species, including *Acacia longifolia*, *Acacia melanoxylon* and *Acacia saligna* in South Africa. The mass of *D. dielsi* galls and adult midges and potential fecundity of the females were about the same on *A. cyclops* and on *A. melanoxylon* but were lower on *A. longifolia* and even lower on *A. saligna*. Cross-over experiments demonstrated that adults emerging from each host-plant species were capable of

mating and producing viable progeny, confirming that these plants are within the fundamental host range of *D. dielsi* and that they have become incorporated into the realized host range of the midge in South Africa. In contrast to *A. cyclops*, these three alternate hosts each have relatively brief flowering periods which precede that of *A. cyclops*. This, in combination with the emergence pattern displayed by *D. dielsi*, seems to play a role in the extent to which the alternate hosts are utilized by *D. dielsi*. All indications are that *D. dielsi* will be of no significance on any species other than *A. cyclops*.

Keywords: Gall midges; Cecidomyiidae; Biological control; Invasive Australian acacias; Fundamental host range; Realized host range; Flowering phenology

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An integrated approach for biological control of crown rot of banana was studied. The efficacy of three control measures, applied alone or in various combinations, was evaluated under conditions highly conducive to the development of crown rot (artificial inoculation of *Colletotrichum musae* at 10^4 conidia/ml). The studied measures were: application of an antagonistic yeast (*Candida oleophila* strain O at 1.10^7 cfu/ml), treatment with 2% (w/v) calcium chloride, and modified atmosphere packaging of fruit (MAP) in non-perforated polyethylene bags. *C. oleophila* was able to grow under MAP, maintaining a large population (7.10^6 to 7.10^7 cfu/g crown) throughout the 13 days of storage. Both treatment with the antagonistic yeast and storage under MAP, applied separately, reduced crown rot significantly (by 22% and 20%, respectively, as compared to untreated controls). The effect of the yeast was the same whether it was produced in Petri dishes or in a fermentor. Calcium chloride treatment alone had no effect on *C. musae*. The antagonistic yeast showed a 16% higher biocontrol activity (from 26% to 42%) when applied together with 2% (w/v) calcium chloride, and the presence of this adjuvant made it possible to achieve the same protective effect with a lower yeast concentration. The highest efficacy (53%) was achieved by the combination of the three alternatives means of control and a synergistic relation has been detected between the yeast, calcium chloride and MAP. Considering the severe conditions of screening, the consistency of the results obtained in this study indicates that the integrated strategy has great potential for control of crown rot of banana under commercial conditions.

Keywords: Banana; *Musa*; *Colletotrichum musae*; Crown rot; Biological control; *Candida oleophila*; Calcium chloride; Modified atmosphere packaging

Yifei Wang[a], Ting Yu[a], Jindan Xia[a], Dasheng Yu[b], Jun Wang[a] and Xiaodong Zheng[a]. ([a] Department of Food Science and Nutrition, Zhejiang University, Hangzhou 310029, People's Republic of China, [b] Vocational and Technical College of Laiwu, Laiwu 271100, People's Republic of China). **Biocontrol of postharvest gray mold of cherry tomatoes with the marine yeast *Rhodosporidium paludigenum*. Biological Control, Volume 53(2) (2010): 178-182**

The marine yeast *Rhodosporidium paludigenum* was tested for its biocontrol efficacy in reducing postharvest gray mold of cherry tomatoes caused by *Botrytis cinerea in vitro* and *in vivo* tests. A previously unreported gray mold antagonist, *R. paludigenum*, significantly reduced disease incidence on cherry tomatoes. The results showed that suspensions containing 1×10^8 CFU/ml washed cells of *R. paludigenum* provided the highest levels of inhibition of gray mold, while the unwashed cell suspension provided less protection against the disease after 5 days at 25 °C. Increasing concentrations of *R. paludigenum* influenced decay incidence in cherry tomato wounds: the disease incidence decreased as the concentration of *R. paludigenum* was increased. Moreover, in the small-scale experiment, the incidence of gray mold on cherry tomatoes treated with *R. paludigenum* was 14.7% and 46.3% of the control at 15 °C after 10 days, respectively. In cherry tomato wounds, the population of *R. paludigenum* reached peak levels 72 h after inoculation, and then began to decline slowly. In conclusion, our results showed that competition for nutrients may have an important role in biological control of gray mold of cherry tomatoes with *R. paludigenum*.

Keywords: *Botrytis cinerea*; *Rhodosporidium paludigenum*; Cherry tomato; Marine antagonist; Biological control

Paola Pereira[a, 1], Andrea Nesci[a, 2], Carlos Castillo[b] and Miriam Etcheverry[a, 2].. ([a]Laboratorio de Ecología Microbiana, Departamento de Microbiología e Inmunología, Facultad de Ciencias Exactas, Físico-Químicas y Naturales, 5800 Rio Cuarto, Córdoba, Argentina, [b] Departamento de Producción Vegetal, Facultad de Agronomía y Veterinaria, Universidad Nacional de Río Cuarto, Ruta Nacional 36 Km 601, 5800 Río Cuarto, Córdoba, Argentina). **Impact of bacterial biological control agents on fumonisin B_1 content and *Fusarium verticillioides* infection of field-grown maize. Biological Control, Volume 53(3) (2010): 258-266**

The effects of four bacterial biocontrol agents on *Fusarium verticillioides* infection and fumonisin accumulation in the maize agroecosystem were evaluated in a 2-year field study. The antagonistic abilities of the four agents were examined following two application techniques consisting of inoculating seeds during pre-sowing and maize ears at flowering. Seed inoculation with *F. verticillioides* and co-inoculation with this fungus and each of the four agents were also examined. Treatment effects on maize performance were also assessed through determination of the number of plants per hectare, kernels yield (kg ha^{-1}) and kernel–cob relations. *F. verticillioides* infection and fumonisin B_1 contents were determined in kernels of physiologically mature maize plants at harvest time. Maize yield remained unchanged with respect to controls in both field surveys; however significantly higher FB_1 contents were obtained after inoculation of seeds with *F. verticillioides* M7075. Seed treatment with *Bacillus amyloliquefaciens* and *Enterobacter hormaechei* reduced the infection by the fungus and FB_1 contents both years of the study while ear inoculation produced highly variable results. The number of colony forming units of *F. verticillioides* obtained from harvested maize kernels was positively correlated with fumonisin B_1 content; however none of these parameters showed significant correlation with kernel yield. The overall results suggest that in years conducive for *F. verticillioides* infection and fumonisin production, seed treatment with *Bacillus amyloliquefaciens* and *Enterobacter hormaechei* may improve quality of maize grains obtained at harvest by reducing toxin content.

Keywords: Biocontrol agents; *Fusarium verticillioides*; Fumonisin B_1; Maize

Khalil I. Al-Mughrabi[a]. ([a] New Brunswick Department of Agriculture and Aquaculture, 39 Barker Lane, Wicklow, New Brunswick, Canada E7L 3S4). Biological control of Fusarium dry rot and other potato tuber diseases using *Pseudomonas fluorescens* and *Enterobacter cloacae*. Biological Control, Volume 53(3) (2010): 280-284

Dry rot caused by various species of *Fusarium* is a disease of significant importance in potatoes. Field trials were conducted in 2005 and 2006 in New Brunswick, Canada to assess the efficacy of *Pseudomonas fluorescens* and *Enterobacter cloacae* applied as a seed treatment in suppressing Fusarium dry rot of potato (*Solanum tuberosum* L.) 'Russet Burbank' under field conditions. In 2005, the trial consisted of five treatments namely (1) non-treated, non-inoculated control; (2) non-treated control inoculated with *Fusarium sambucinum*; (3) seed inoculated with *F. sambucinum* and treated with *P. fluorescens*; (4) seed inoculated with *F. sambucinum* and treated with *E. cloacae*; and (5) seed inoculated with *F. sambucinum* and treated with the fungicide fludioxonil. In 2006, a mustard meal treatment was added. After harvest, tubers were assessed for disease severity of dry rot in addition to other tuber diseases, including silver scurf (*Helminthosporium solani*), and common scab (*Streptomyces scabiei*). In addition, tubers were graded and assessed for total yield, tuber size, tuber number, and tuber weight. Significant reduction in dry rot severity was obtained with all treatments compared to the non-treated control inoculated with *Fusarium sambucinum*. The highest dry rot reduction averaged over the two years of the study was for the fludioxonil treatment (55.7%) followed by the treatment with mustard meal (47.5%; 2006 only), *Pseudomonas fluorescens* (35%) and *Enterobacter cloacae* (26.5%). All treatments significantly reduced the severity of common scab and silver scurf compared to the non-treated, non-inoculated control. In both years, on an average, seed treated with *P. fluorescens* and *E. cloacae* produced higher total number of tubers. Both total and marketable tuber yields were significantly higher for the *E. cloacae* treatment compared to the non-treated, inoculated control. The results of this study suggest that *P. fluorescens*, *E. cloacae* and mustard meal are viable options for controlling potato tuber diseases along with fludioxonil. This is the first study to investigate the effect of these bacteria on potato diseases under field settings.

Keywords: Biological control; *Enterobacter cloacae*; Fludioxonil; *Fusarium sambucinum*; *Helminthosporium solani*; Mustard meal; *Pseudomonas fluorescens*; *Streptomyces scabiei*

C.J. Wijesinghe[a], R.S. Wilson Wijeratnam[a], J.K.R.R. Samarasekara[b] and R.L.C. Wijesundera[c]. ([a] Post Harvest Technology Laboratory, Industrial Technology Institute, 363, Bauddhaloka Mawatha, Colombo 07, Sri Lanka, [b] Herbal Technology Laboratory, Industrial Technology Institute, 363, Bauddhaloka Mawatha, Colombo 07, Sri Lanka, [c] Department of Plant Sciences, University of Colombo, Colombo 03, Sri Lanka). Biological control of *Thielaviopsis paradoxa* on pineapple by an isolate of *Trichoderma asperellum*. Biological Control, Volume 53(3) (2010): 285-290

An isolate of *Trichoderma asperellum* was tested for its antagonistic activity against *Thielaviopsis paradoxa*, the causal agent of black rot of pineapple, *Anansas comosus*. Results of our investigation clearly demonstrated antagonistic activity of *T. asperellum* against *Th. paradoxa*. The antagonistic activity was mainly due to coil formation around the pathogen hyphae by *T. asperellum*. The activity was found to be fungicidal. Pineapple fruits inoculated with 10^5 conidia/mL of *Th. paradoxa*, followed by an application of a formulation containing spores of *T. asperellum* within 10 and 30 min after inoculation, were free of disease when stored at 28 °C for 7 days. Fruits treated with the blank formulation (formulation without conidia) and

the control fruits showed characteristic symptoms of black rot disease. Black rot symptoms also were observed on fruits that were inoculated with *Th. paradoxa* and held as inoculated controls under similar storage conditions. Chemical parameters namely; pH and titratable acidity among treated, untreated (controls) and fruits treated with a blank formulation showed no significant difference. The data suggest that a formulation of *T. asperellum* might be an alternate and ecofriendly method for the control of black rot disease of Pineapple.

Keywords: *Trichoderma asperellum*; Biocontrol formulations; Pineapple (*Ananas comosus*) black rot disease

Dolors Sant[a], Eva Casanova[b], Guillem Segarra[a], Manuel Avilés[c], Mário Reis[d] and M. Isabel Trillas[a]. ([a] Universitat de Barcelona, Departament de Biologia Vegetal, Facultat de Biologia, Avgda. Diagonal 645, E-08028 Barcelona, Spain, [b] Biocontrol Technologies, S.L. Parc Científic de Barcelona, Baldiri Reixac 15-21, E-08028 Barcelona, Spain, [c] Universidad de Sevilla, Departamento de Ciencias Agroforestales, E.U.I.T.A., Ctra. Utrera, Km. 1, s/n, E-41013 Sevilla, Spain, [d] Universidade do Algarve, Faculdade de Ciências e Tecnologias, Campus de Gambelas, 8005-139 Faro, Portugal). Effect of *Trichoderma asperellum* strain T34 on Fusarium wilt and water usage in carnation grown on compost-based growth medium. Biological Control, Volume 53(3) (2010): 291-296

Suppressive composts are viable alternative growth media for the biological control of plant diseases. To adapt the physical properties of these composts to growth conditions, formulation is sometimes required and consequently their suppressive capacity is reduced. Green house experiments show that plant growth medium based on grape marc compost (compost:peat 1:1, v/v) amended with the biological control agent *Trichoderma asperellum* strain T34, restores composts' suppressive capacity against Fusarium wilt of carnation compared with the non-amended medium. Standard chemicals (tocloflos-methyl 50% and captan 85%) used in the cultivation of carnation crops did not improve the suppressive capacity of the growth media or the T34 populations. However, they impaired the stomatal conductance of plants and increased the populations of the pathogen (interaction chemical and T34). Water use measurements of carnation plants, namely stomatal conductance, transpiration and daily water uptake, correlated negatively with Fusarium wilt disease. Plants in T34-amended growth media had higher water demand, regardless of the treatment, which implied their growth could be enhanced by increasing fertirrigation, compared to plants non-amended with T34. In conclusion, the amendment of formulated grape marc compost with *T. asperellum* T34 improves the suppressive capacity of this growth medium against Fusarium wilt in carnation. It is a better alternative for plant and disease control than the standard chemicals used.

Keywords: Biological control agent; *Dianthus caryophyllus*; *Fusarium oxysporum* f.sp. *dianthi*; Grape marc compost; Transpiration; Stomatal conductance; Water uptake

Yanghui Xiong[1] and Yu Liu[1]. ([1]Division of Environmental and Water Resources Engineering, School of Civil and Environmental Engineering, Nanyang Technological University, 50 Nanyang Avenue, Singapore, 639798, Singapore). Biological control of microbial attachment: a promising alternative for mitigating membrane biofouling. Applied Microbiology and Biotechnology, Volume 86(3) (2010): 825-837

Microbial attachment to a solid surface is a universal phenomenon occurring in both natural and engineering systems and is responsible for various types of biofouling. Membrane systems have been widely applied in drinking water production, wastewater reuse, and seawater desalination. However, membrane biofouling is the bottleneck that limits the development of membrane systems. In this review, some biological control strategies of microbial attachment which would have great potential in alleviating membrane biofouling are discussed, including inhibition of quorum sensing system, nitric oxide-induced biofilm dispersal, enzymatic disruption of extracellular polysaccharides, proteins, and DNA, inhibition of microbial attachment by energy uncoupling, use of cell wall hydrolases, and disruption of biofilm by bacteriophage. It appears that biological control of microbial attachment would be a novel and promising alternative for mitigating membrane biofouling and would be a new research niche that deserves further study.

Keywords: Microbial attachment - Membrane fouling - Quorum sensing - Autoinducers - Nitric oxide - EPS - eDNA - Energy uncoupling - Cell wall hydrolases – Bacteriophage

Zhen-Ming Chi[1], Guanglei Liu[1], Shoufeng Zhao[1], Jing Li[1] and Ying Peng[1]. ([1]Unesco Chinese Center of Marine Biotechnology, Ocean University of China, Yushan Road, No. 5, Qingdao, China). **Marine yeasts as biocontrol agents and producers of bio-products. Applied Microbiology and Biotechnology, Volume 86(5) (2010): 1227-1241**

As some species of marine yeasts can colonize intestine of marine animals, they can be used as probiotics. It has been reported that β-glucans from marine yeast cells can be utilized as immuno-stimulants in marine animals. Some siderophores or killer toxins produced by marine yeasts have ability to inhibit growth of pathogenic bacteria or kill pathogenic yeasts in marine animals. The virulent factors from marine pathogens can be genetically displayed on marine yeast cells, and the yeast cells displaying the virulent factors can stimulate marine animals to produce specific antibody against the pathogens. Some marine yeast cells are rich in proteins and essential amino acids and can be used in nutrition for marine animals. The marine yeast cells rich in lipid can be used for biodiesel production. Recently, it has been reported that some strains of *Yarrowia lipolytica* isolated from marine environments can produce nanoparticles. Because many marine yeasts can remove organic pollutants and heavy metals, they can be applied to remediation of marine environments. It has been shown that the enzymes produced by some marine yeasts have many unique properties and many potential applications.

Keywords: Probiotics - Marine yeasts - Industrial enzymes - Vaccine - Siderophore - Bio-products

Prasun K. Mukherjee[1,2] and Charles M. Kenerley[1*] (Department of Plant Pathology and Microbiology, Texas A&M University, College Station, Texas 77843,[1] Nuclear Agriculture and Biotechnology Division, Bhabha Atomic Research Center, Trombay, Mumbai 400085, India[2]). **Regulation of Morphogenesis and Biocontrol Properties in *Trichoderma virens* by a VELVET Protein, Vel1[▽,†]. Applied and Environmental Microbiology, Vol. 76(7) (2010): 2345-2352**

Mycoparasitic strains of *Trichoderma* are applied as commercial biofungicides for control of soilborne plant pathogens. Although the majority of commercial biofungicides are *Trichoderma* based, chemical pesticides, which are ecological and environmental hazards, still dominate the market. This is because biofungicides are not as effective or consistent as chemical fungicides. Efforts to improve these products have been limited by a lack of understanding of the genetic

regulation of biocontrol activities. In this study, using gene knockout and complementation, we identified the VELVET protein Vel1 as a key regulator of biocontrol, as well as morphogenetic traits, in *Trichoderma virens*, a commercial biocontrol agent. Mutants with mutations in *vel1* were defective in secondary metabolism (antibiosis), mycoparasitism, and biocontrol efficacy. In nutrient-rich media they also lacked two types of spores important for survival and development of formulation products: conidia (on agar) and chlamydospores (in liquid shake cultures). These findings provide an opportunity for genetic enhancement of biocontrol and industrial strains of *Trichoderma*, since Vel1 is very highly conserved across three *Trichoderma* species.

Minakshi Grover[1] , Lata Nain[2], Shashi Bala Singh[3] and Anil Kumar Saxena[2]. ([1]Division of Crop Sciences, Central Research Institute for Dryland Agriculture, Hyderabad, 50059, India, [2]Division of Microbiology, Indian Agricultural Research Institute, New-Delhi, 110012, India, [3]Division of Agricultural Chemicals, Indian Agricultural Research Institute, New-Delhi, 110012, India). Molecular and Biochemical Approaches for Characterization of Antifungal Trait of a Potent Biocontrol Agent *Bacillus subtilis* RP24. Current Microbiology, Volume 60(2) (2010): 99-106

Bacillus subtilis strain RP24, isolated from rhizoplane of field grown pigeon pea, exhibited in vitro antagonism against a wide range of phytopathogenic fungi. An attempt was made to partially purify and characterize the diffusible antifungal metabolite/s produced by the strain RP24 and its negative mutant (NM) in potato dextrose medium. High performance liquid chromatography (HPLC) of partially purified extract of RP24 showed the presence of lipopeptide antibiotic iturin as a major peak that was comparable to that of standard iturin A (5.230 min) from Sigma–Aldrich whereas the corresponding peak was absent in extract of NM. The structure was further confirmed by liquid chromatographic mass spectrometric (LCMS) analysis as iturin A. LCMS analysis also showed the presence of surfactin and fengycin besides iturin A. Amplification of the *lpa*-14 (encodes the 4′-phosphopantetheinyl transferase required for the maturation of template enzyme of iturin A) and *itu*D (encodes a putative malonyl coenzyme A transacylase, whose disruption results in a specific deficiency in iturin A production) genes of iturin operon of strain RP24 was carried out and the sequences obtained were compared with the existing database of NCBI. The sequences of *lpa*-14 and *itu*D gene of RP24 showed 98% and 97% homology with *lpa*-14 and *itu*D genes of *B. subtilis* in the existing database. The results indicated that strain RP24 harbors iturin operon in its genome and a chemical mutation in this operon might have resulted in loss of antifungal activity in the negative mutant.

Biodegradation

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Methyl iso-butyl ketone (MIBK) is a widely used volatile organic compound (VOC) which is highly toxic in nature and has significant adverse effects on human beings. The present study

deals with the removal of MIBK using biodegradation by an acclimated mixed culture developed from activated sludge. The biodegradation of MIBK is studied for an initial MIBK concentration ranging from 200–700 mg l^{-1} in a batch mode of operation. The maximum specific growth rate achieved is 0.128 h^{-1} at 600 mg $l^{-1}$of initial MIBK concentration. The kinetic parameters are estimated using five growth kinetic models for biodegradation of organic compounds available in the literature. The experimental data found to fit well with the Luong model ($R^2 = 0.904$) as compared to Haldane model ($R^2 = 0.702$) and Edward model ($R^2 = 0.786$). The coefficient of determination (R^2) obtained for the other two models, Monod and Powell models are 0.497 and 0.533, respectively. The biodegradation rate found to follow the three-half-order kinetics and the resulting kinetic parameters are reported.

Keywords: Biodegradation - Acclimated mixed culture - Biomass - Methyl iso-butyl ketone - Growth kinetics - Inhibition - Rate kinetics

Priyangshu Manab Sarma[1], Prem Duraja[2], Shilpanjali Deshpande[3] and Banwari Lal[4]. ([1]Microbial Biotechnology, The Energy and Resources Institute, Habitat Place, Lodhi Road, New Delhi, 110003, India, [2]Division of Agricultural Chemicals, Indian Agricultural Research Institute, PUSA Road, New Delhi, 110012, India, [3]Center for Bioresource and Biotechnology, TERI University, Vasant Kunj, New Delhi, 110070, India, [4]Environmental and Industrial Biotechnology, The Energy and Resources Institute, Habitat Place, Lodhi Road, New Delhi, 110 003, India). **Degradation of pyrene by an enteric bacterium, *Leclercia adecarboxylata* PS4040. Biodegradation, Volume 21(1) (2010): 59-69**

A newly discovered enteric bacterium *Leclercia adecarboxylata* PS4040, isolated from oily sludge contaminated soil sample was reported for degradation of polycyclic aromatic hydrocarbons (Appl Environ Microbiol 70:3163–3166, *2004a*). This strain could degrade 61.5% of pyrene within 20 days when used as sole source of carbon and energy. The time course degradation experiment detected several intermediate products and the metabolites were identified by gas chromatography mass spectrometry analysis. Metabolite I was the detected on the 5th day and was identified as 1-hydroxypyrene and was detected till 10th day. Metabolite II which was detected on 10th day was identified as 1,2-phenanthrenedicarboxylic acid. Metabolite III and Metabolite IV were identified as 2-carboxy benzaldehyde and *ortho*-phthalic acid, respectively and were detected in the culture broth on 10th and 15th day. 1,2-benzene diol (catechol) was the fifth metabolite detected in the culture extracts on the 15th day and was subsequently reduced on day 20. Identification of Metabolite I as 1-hydroxypyrene was further investigated as this intermediate was not previously reported as a ring oxidation product for degradation of pyrene by bacterial strains. Purification by preparative high performance liquid chromatography and nuclear magnetic resonance spectroscopy, confirmed the identification of Metabolite I as 1-hydroxypyrene. *L. adecarboxylata* PS4040 could also use 1-hydroxypyrene as a sole source of carbon and energy. Thus a probable pathway for degradation of pyrene by enteric bacterium is proposed in this study, with 1-hydroxypyrene as initial ring oxidation product.

Keywords: Pyrene degradation - Enteric bacteria - *Leclercia adecarboxylata*

Chunyu Yang[1], Yang Li[1], Kun Zhang[1], Xia Wang[1], Cuiqing Ma[1], Hongzhi Tang[2] and Ping Xu[1, 2]. ([1]State Key Laboratory of Microbial Technology, Shandong University, 250100 Jinan, People's Republic of China, [2]MOE Key Laboratory of Microbial Metabolism, School of Life Sciences and Biotechnology, Shanghai Jiao Tong University,

200240 Shanghai, People's Republic of China). Atrazine degradation by a simple consortium of *Klebsiella* sp. A1 and *Comamonas* sp. A2 in nitrogen enriched medium. Biodegradation, Volume 21(1) (2010): 97-105

A simple consortium consisted of two members of *Klebsiella* sp. A1 and *Comamonas* sp. A2 was isolated from the sewage of a pesticide mill in China. One member of *Klebsiella* sp. A1 is a novel strain that could use atrazine as the sole carbon and nitrogen source. The consortium showed high atrazine-mineralizing efficiency and about 83.3% of 5 g l^{-1} atrazine could be mineralized after 24 h degradation. Contrary to many other reported microorganisms, the consortium was insensitive to some nitrogenous fertilizers commonly used, not only in presence of 200 mg l^{-1} atrazine but also in 5 g l^{-1} atrazine mediums. After 24 h incubation, 200 mg l^{-1} atrazine was completely mineralized despite of the presence of urea, $(NH_4)_2CO_3$ and $(NH_4)_2HPO_4$ in the medium. Very minor influence was observed when NH_4Cl was added as additional nitrogen source. Advantages of the simple consortium, high mineralizing efficiency and insensitivity to most of exogenous nitrogen sources, all suggested application potential of the consortium for the bioremediation of atrazine-contaminated soils and waters.

Keywords: Atrazine - Bioremediation - Consortium - *Klebsiella* - Nitrogen insensitivity

Jie Xu[1] and Qian Yang[1]. ([1]Department of Life Science and Engineering, Harbin Institute of Technology, 150001 Harbin, China). Isolation and characterization of rice straw degrading *Streptomyces griseorubens* C-5. Biodegradation, Volume 21(1) (2010): 107-116

To reutilize rice straw generated during the agricultural production process, the actinomycete strain C-5 was isolated from soil that was under the stook for several years in the Heilongjiang province of China by using multiple selective culture media. Strain C-5 was identified as *Streptomyces griseorubens* by China General Microbiological Culture Collection Center (CGMCC) through morphological and physiological characterization combined with the result of 16S rRNA gene sequence and data analysis. This strain has simultaneous cellulase, laccase, peroxidase, xylanase and pectinase activity. The various chemical composition of rice straw were determined during fermentation process. Simultaneously the biodegradation process of rice straw stem was observed by scanning electron microscope (SEM). It is predicted that strain C-5 could decompose rice straw effectively and had promising prospects of being applied in improving the resource utilization of rice straw.

Keywords : Biodegradation - Rice straw - Lignocellulose - *Streptomyces griseorubens* - Scanning electron microscope

Xin Cheng[1], Lin Huang[1], Xiao-rong Tu[1] and Kun-tai Li[1]. ([1]Nanchang Key Laboratory of Applied Fermentation Technology, Jiangxi Agricultural University, 330045 Nanchang, China). Medium optimization for the feather-degradation by *Streptomyces fradiae VarS-221* using the response surface methodology. Biodegradation, Volume 21(1) (2010): 117-122

In order to accelerate biodegradation of feather into more amino acids, the fermentation medium of feather-biodegrading *Streptomyces fradiae Var S-221* was optimized in this paper. In the first optimization step, the effects of feather powder, beet molasses, $(NH_4)_2SO_4$ and KH_2PO_4 on amino acids formation were evaluated by using full factorial design. The results showed that

feather powder and $(NH_4)_2SO_4$ had significant and positive effects on feather-biodegradation into amino acids. Then, the method of the steepest ascent was used to access the optimal region of the two significant factors. In the third step, the concentration of feather powder and $(NH_4)_2SO_4$ were further optimized with central composite design and response surface analysis. As a result, the composition of the optimal medium for *S. fradiae Var S-221* fermentation were as follows (g/100 ml): feather powder, 19.504; beet molasses, 4.0; $(NH_4)_2SO_4$, 1.467; KH_2PO_4, 0.3; $MgSO_4$, 0.15; $FeSO_4$, 0.001; $ZnSO_4$, 0.0001; and $MnSO_4$, 0.0001. Using this optimal fermentation medium, the amino acids concentration was increased from 4.61 to 6.13 g/100 ml.

Keywords: Amino acid - Feather-biodegradation - Medium optimization - *Streptomyces fradiae Var S-221* - Response surface methodology

J. Jacob Parnell[1,4], Vincent J. Denef[2], Joonhong Park[3], Tamara Tsoi[1] and James M. Tiedje[1]. ([1]Center for Microbial Ecology and Crop and Soil Science, Michigan State University, East Lansing, MI 48823, USA, [2]Department of Earth and Planetary Sciences, University of California Berkeley, Berkeley, CA, USA, [3]School of Civil and Environmental Engineering, Yonsei University, Seoul, 120-749, Republic of Korea, [4]*Present address:* Center for Integrated BioSystems and Department of Biology, Utah State University, Logan, UT, USA). **Environmentally relevant parameters affecting PCB degradation: carbon source- and growth phase-mitigated effects of the expression of the biphenyl pathway and associated genes in *Burkholderia xenovorans* LB400. Biodegradation, Volume 21(1) (2010): 147-156**

The principal means for microbial degradation of polychlorinated biphenyls (PCBs) is through the biphenyl pathway. Although molecular aspects of the regulation of the biphenyl pathway have been studied, information on environmental facets such as the effect of alternative carbon sources on (polychlorinated) biphenyl degradation is limited. Here we explore the effect of environmental conditions (e.g., carbon source and growth phase) on the variation in PCB degradation profiles of *Burkholderia xenovorans* LB400. Genome-wide expression patterns reveal 25 genes commonly up-regulated during PCB degradation and growth on biphenyl to be upregulated in the transition to stationary phase (relative to growth on succinate) including two putative detoxification pathways. Quantitative reverse transcription PCR (Q-RT-PCR) analysis of the upper biphenyl pathway (*bphA*, *bphD*, and *bphR1*), and detoxification genes in response to environmental conditions suggest associated regulation of the biphenyl pathway and chloroacetaldehyde dehydrogenase. The response of genes in the upper biphenyl pathway to carbon source competition and growth phase reveals inhibition of the biphenyl pathway by PCBs. Although PCBs are not degraded during growth on succinate with PCBs, expression data indicate that the biphenyl pathway is induced, suggesting that post-transcriptional regulation or active transport of biphenyl maybe limiting PCB degradation. Identification of the involvement of peripheral pathways in degradation of PCBs is crucial to understanding PCB degradation in an environmental context as bacteria capable of biodegradation experience a range of carbon sources and growth phases.

Keywords: Polychlorinated biphenyl degradation - Cometabolism - Biphenyl degradation pathway - *Burkholderia xenovorans* LB400

Tadashi Toyama[1], Noritaka Maeda[1], Manabu Murashita[1], Yong-Cheol Chang[1] and Shintaro Kikuchi[1]. ([1]Division of Applied Sciences, Muroran Institute of Technology, 27-1 Mizumoto, Muroran 050-8585, Japan). **Isolation and characterization of a novel 2-*sec*-**

butylphenol-degrading bacterium *Pseudomonas* sp. strain MS-1. Biodegradation, Volume 21(2) (2010): 157-165

A novel bacterium capable of utilizing 2-*sec*-butylphenol as the sole carbon and energy source, *Pseudomonas* sp. strain MS-1, was isolated from freshwater sediment. Within 30 h, strain MS-1 completely degraded 1.5 mM 2-*sec*-butylphenol in basal salt medium, with concomitant cell growth. A pathway for the metabolism of 2-*sec*-butylphenol by strain MS-1 was proposed on the basis of the identification of 3 internal metabolites—3-*sec*-butylcatechol, 2-hydroxy-6-oxo-7-methylnona-2,4-dienoic acid, and 2-methylbutyric acid—by gas chromatography-mass spectrometry analysis. Strain MS-1 degraded 2-*sec*-butylphenol through 3-*sec*-butylcatechol along a *meta*-cleavage pathway. Degradation experiments with various alkylphenols showed that the degradability of alkylphenols by strain MS-1 depended strongly on the position (*ortho meta* = *para*) of the alkyl substitute, and that strain MS-1 could degrade 2-alkylphenols with various sized and branched alkyl chain (*o*-cresol, 2-ethylphenol, 2-*n*-propylphenol, 2-isopropylphenol, 2-*sec*-butylphenol, and 2-*tert*-butylphenol), as well as a dialkylphenol (namely, 6-*tert*-butyl-*m*-cresol).

Keywords: 2-*sec*-Butylphenol - *ortho*-Substituted alkylphenol - Biodegradation - *meta*-Cleavage pathway

San-Lang Wang[1, 2], Tao-Jen Chang[1] and Tzu-Wen Liang[2]. ([1]Graduate Institute of Life Sciences, Tamkang University, Taipei, 251, Taiwan, [2]Life Science Development Center, Tamkang University, Taipei, 251, Taiwan). **Conversion and degradation of shellfish wastes by *Serratia* sp. TKU016 fermentation for the production of enzymes and bioactive materials. Biodegradation, Volume 21(3) (2010): 321-333**

A chitosanase and a protease were purified from the culture supernatant of *Serratia* sp. TKU016 with shrimp shell as the sole carbon/nitrogen source. The molecular masses of the chitosanase and protease determined by SDS–PAGE were approximately 65 and 53 kDa, respectively. The chitosanase was inhibited completely by Mn^{2+}, but the protease was enhanced by all of tested divalent metals. The optimum pH, optimum temperature, pH stability, and thermal stability of the chitosanase and protease were (pH 7, 50°C, pH 6–7, <50°C) and (pH 8–10, 40°C, pH 5–10, <50°C), respectively. SDS (2 mM) had stimulatory effect on TKU016 protease activity. The result demonstrates that TKU016 protease is SDS-resistant protease and probably has a rigid structure. Besides, TKU016 culture supernatant (2% SPP) incubated for 2 days has the highest antioxidant activity, the DPPH scavenging ability was about 76%. With this method, we have shown that shrimp shell wastes can be utilized and it's effective in the production of enzymes, antioxidants, peptide and reducing sugar, facilitating its potential use in biological applications and functional foods.

Keywords : Chitosanase - Protease - Antioxidant activity - *Serratia* sp. - Shrimp shell wastes

Yaohui Bai[1], Qinghua Sun[1], Cui Zhao[1], Donghui Wen[1] and Xiaoyan Tang[1]. ([1]College of Environmental Sciences and Engineering, Peking University, Beijing, 100871, People's Republic of China). Quinoline biodegradation and its nitrogen transformation pathway by a *Pseudomonas* sp. Strain. Biodegradation, Volume 21(3) (2010): 335-344

A *Pseudomonas* sp. strain, which can utilize quinoline as its sole carbon, nitrogen and energy source, was isolated from activated sludge in a coking wastewater treatment plant. Quinoline can be degraded via the 8-hydroxycoumarin pathway. We quantified the first two organic intermediates of the biodegradation, 2-hydroxyquinoline and 2,8-dihydroxyquinoline. We tracked the transformation of the nitrogen in quinoline in two media containing different C/N ratios. At least 40.4% of the nitrogen was finally transformed into ammonium when quinoline was the sole C and N source. But addition of an external carbon source like glucose promoted the transformation of N from NH_3 into NO_3^-, NO_2^-, and then to N_2. The product analysis and gene characteristics indicated that the isolate accomplished heterotrophic nitrification and aerobic denitrification simultaneously. The study also demonstrated that quinoline and its metabolic products can be eliminated if the C/N ratio is properly controlled in the treatment of quinoline-containing wastewater.

Keywords: *Pseudomonas* sp. - Quinoline - Biodegradation - Nitrification - Denitrification

Nucleotide sequence accession number The accession numbers of the isolates 16S rRNA gene, *nirS*, and *nosZ* on GenBank are EU266621, FJ393272 and FJ393273.

Itza Mendoza-Sanchez[1], Robin L. Autenrieth[2], Thomas J. McDonald[3] and Jeffrey A. Cunningham[4]. ([1]Escuela Superior de Ingeniería y Arquitectura, Instituto Politécnico Nacional, Mexico City, Mexico, [2]Department of Civil Engineering, Texas A & M University, College Station, TX 77843, USA, [3]School of Rural Public Health, Texas A & M University, College Station, TX 77843, USA, [4]Department of Civil and Environmental Engineering, University of South Florida, Tampa, FL 33620, USA). Effect of pore velocity on biodegradation of *cis*-dichloroethene (DCE) in column experiments. Biodegradation, Volume 21(3) (2010): 365-377

Column experiments were conducted to evaluate the effect of pore velocity on the extent of biodegradation of *cis*-dichloroethene (*cis*-DCE) during transport in porous media. Columns were filled with homogeneous glass beads and inoculated with a culture capable of complete dechlorination of tetrachloroethene to ethene. A constant concentration of *cis*-DCE was maintained in the columns' influent. Three different pore velocities were tested in duplicate, subjecting each column to a constant velocity. At high flow velocity, degradation of *cis*-DCE to ethene was nearly complete within the residence time of the columns. However, at medium and low flow velocities, incomplete dechlorination was observed. After 7 weeks, DNA was harvested from the columns to determine differences in the microbial populations. Results suggest that *Dehalococcoides* sp. were present in higher quantities in the high-velocity columns, consistent with the observed dechlorination. These results suggest that, at contaminated groundwater sites, heterogeneity of groundwater velocity may be one factor that contributes to heterogeneous distribution of biological activity.

Keywords : Bioremediation - Chlorinated solvents - PCE - TCE - Vinyl chloride - KB-1

Jyoti P. Jadhav[1], Swapnil S. Phugare[1], Rhishikesh S. Dhanve[1] and Shekhar B. Jadhav[1] ([1]Department of Biochemistry, Shivaji University, Kolhapur, 416 004, India). **Rapid biodegradation and decolorization of Direct Orange 39 (Orange TGLL) by an isolated bacterium *Pseudomonas aeruginosa* strain BCH. Biodegradation, Volume 21(3) (2010): 453-463**

A newly isolated novel bacterium from sediments contaminated with dyestuff was identified as *Pseudomonas aeruginosa* strain BCH by 16S rRNA gene sequence analysis. The bacterium was extraordinarily active and operative over a wide rage of temperature (10–60°C) and salinity (5–6%), for decolorization of Direct Orange 39 (Orange TGLL) at optimum pH 7. This strain was capable of decolorizing Direct Orange 39; 50 mg l^{-1} within 45 ± 5 min, with 93.06% decolorization, while maximally it could decolorize 1.5 g l^{-1} of dye within 48 h with 60% decolorization. Analytical studies as, UV–Vis spectroscopy, FTIR, HPLC were employed to confirm the biodegradation of dye and formation of new metabolites. Induction in the activities of lignin peroxidases, DCIP reductase as well as tyrosinase was observed, indicating the significant role of these enzymes in biodegradation of Direct Orange 39. Toxicity studies with *Phaseolus mungo* and *Triticum aestivum* revealed the non-toxic nature of degraded metabolites.

Keywords: Biodegradation - DCIP reductase - Direct Orange 39 - Salinity - Tyrosinase

Naoki Harada[1], Kazuhiro Takagi[2], Koji Baba[2], Kunihiko Fujii[3] and Akio Iwasaki[3]. ([1]Faculty of Agriculture, Niigata University, 2-8050 Ikarashi, Nishi-ku Niigata, 950-2181, Japan, [2]Organochemicals Division, National Institute for Agro-Environmental Sciences, 3-1-3 Kannondai, Tsukuba Ibaraki, 305-8604, Japan, [3]Kowa Research Institute, Kowa Co., Ltd., 1-25-5 Kannondai, Tsukuba Ibaraki, 305-0856, Japan). **Biodegradation of diphenylarsinic acid to arsenic acid by novel soil bacteria isolated from contaminated soil. Biodegradation, Volume 21(3) (2010): 491-499**

Microorganisms capable of degrading diphenylarsinic acid (DPAA) were enriched from contaminated soil using the soil-charcoal perfusion method. Two novel bacterial strains, L2406 and L2413, that can degrade DPAA in a mineral salt medium supplemented with DPAA as the sole carbon source were isolated. Based on comparative morphology, physiology, and comparison of the 16S rRNA gene sequences, both were presumed to be species closely related to *Ensifer adhaerens*. As the metabolites, phenylarsonic acid (PAA) was determined by liquid chromatography-mass spectrometry analysis as well as three unknown peaks all of whose molecular weights were estimated to be 278. The increase of m/z = 16 from DPAA in the unknowns suggests monohydroxylation of DPAA at the 2-, 3- and 4-positions. The ability of strains L2406 and L2413 to degrade DPAA was suppressed in iron insufficient conditions, e.g. less than 7.2 μM iron in the culture medium. These facts strongly suggest the following hypothesis: Monooxygenase works at the initial degradation step of DPAA degradation by the isolates; and direct hydrolysis from DPAA to PAA is not likely to occur. In addition, release of arsenic acid from PAA by strain L2406 was confirmed by liquid chromatography-inductively coupled plasma mass spectrometry. From these results, strain L2406 was considered to be capable of degrading DPAA to arsenic acid via PAA when DPAA was supplied as the sole carbon source.

Keywords: Biodegradation - Diphenylarsinic acid - *Ensifer adhaerens* - LC–MS - Phenylarsonic acid

Hosseini Farzaneh[1]*, Malekzadeh Fereidon[1], Amirmozafari Noor[2] and Ghaemi Naser[3]. ([1]Department of Biology, College of Basic Science, Islamic Azad University Research and Science Tehran branch, Tehran, Iran, [2]Department of microbiology, Iran University of Medical Sciences, Tehran, Iran, [3]Department of Biotechnology, College of Science, Tehran University, Tehran, Iran. *Corresponding author. E-mail: farzaneh953@yahoo.com. Tel: +98021-22641842). **Biodegradation of dodecylbenzene solfonate sodium by *Stenotrophomonas maltophilia* Biofilm. African Journal of Biotechnology, Vol. 9 (1) (2010): 055-062**

Immobilization for microbial cultures has proved to be advantageous in municipal and industrial sewage treatment because of high degradation efficiency and good operational stability. In this survey, a bacterial strain was isolated from activated sludge that utilized branched anionic surfactants (BAS) as a sole carbon source. Identification of isolated strain was from 16S rRNA sequencing method. The immobilized cells on silanized glass beads as support and unmodified surfaces were used for removal of BAS; both types showed effective biodegrading of BAS. The removal rate in silanized surface was about 2 fold of unmodified surface. The result of biodegradation was studied by HPLC method and scanning electron microscope.

Key words: Branched anionic surfactants, biofilm, biodegradation, silanization.

Li Kang[1], Wei Wang[1] and Yoon Y. Lee[1]. ([1]Department of Chemical Engineering, Auburn University, Auburn, AL 36849, USA). **Bioconversion of Kraft Paper Mill Sludges to Ethanol by SSF and SSCF. Applied Biochemistry and Biotechnology, Volume 161(1-8) (2010): 53-66**

Paper mill sludge is a solid waste material composed of pulp residues and ash generated from pulping and paper making processes. The carbohydrate portion of the sludge has chemical and physical characteristics similar to pulp. Because of its high carbohydrate content and well-dispersed structure, the sludges can be biologically converted to value-added products without pretreatment. In this study, two different types of paper mill sludges, primary sludge and recycle sludge, were evaluated as a feedstock for bioconversion to ethanol. The sludges were first subjected to enzymatic conversion to sugars by commercial cellulase enzymes. The enzymatic conversion was inefficient because of interference by ash in the sludges with the enzymatic reaction. The main cause was that the pH level is dictated by $CaCO_3$ in ash, which is two units higher than the pH optimum of cellulase. To alleviate this problem, simultaneous saccharification and cofermentation (SSCF) using cellulase (Spezyme CP) and recombinant *Escherichia coli* (ATCC-55124), and simultaneous saccharification and fermentation (SSF) using cellulase and *Saccharomyces cerevisiae* (ATCC-200062) were applied to the sludges without any pretreatment. Ethanol yields of 75–81% of the theoretical maximum were obtained from the SSCF on the basis of total carbohydrates. The yield from the SSF was also found to be in the range of 74–80% on the basis of glucan. The SSCF and SSF proceeded under stable condition with the pH staying near 5.0, close to the optimum for cellulase. Decrease of pH occurred due to carbonic acid and other organic acids formed during fermentation. The ash was partially neutralized by the acids produced from the SSCF and SSF and acted as a buffer to stabilize the pH during fermentation. When the SSF and SSCF were operated in fed-batch mode, the ethanol concentration in the broth increased from 25.5 and 32.6 g/L (single feed) to 45 and

42 g/L, respectively. The ethanol concentration was limited by the tolerance of the microorganism in the case of SSCF. The ethanol yield in fed-batch operation decreased to 68% for SSCF and 70% for SSF. The high-solids condition in the bioreactor appears to create adverse effects on the cellulase reaction.

Keywords: Paper mill sludges - Ethanol - SSF - SSCF - Bioconversion

Yanna Liang[1], Jemil Yesuf[1] and Zisong Feng[1]. ([1]Department of Civil and Environmental Engineering, Southern Illinois University Carbondale, 1230 Lincoln Dr., Carbondale, IL 62901, USA). Toward Plant Cell Wall Degradation Under Thermophilic Condition: A Unique Microbial Community Developed Originally from Swine Waste. Applied Biochemistry and Biotechnology, Volume 161(1-8) (2010): 147-156

A unique thermophilic microbial community developed initially from swine waste was investigated in this study. Cellulase activities were observed when this community was inoculated to media containing either cellulose or carboxymethylcellulose at 57 °C. Through constructing a clone library for the 16S ribosomal DNA, it was revealed that this community was mainly composed of three genera: *Thermobacillus*, *Brevibacillus*, and *Anoxybacillus*. New findings regarding the thermo- and pH stability of crude cellulases secreted by *Brevibacillus* sp. JXL were presented. Recent study on the growth characteristics of *Anoxybacillus* sp. 527 was discussed.

Keywords: Cellulose - Thermophilic - Cellulase - Microbial community - *Brevibacillus* sp. JXL - *Anoxybacillus* sp. 527

Jenny Rattfelt Nyholm[a], Charlott Lundberg[a] and Patrik L. Andersson[a]. ([a]Department of Chemistry, Umeå University, SE-901 87 Umeå, Sweden). Biodegradation kinetics of selected brominated flame retardants in aerobic and anaerobic soil. Environmental Pollution, Volume 158(6) (2010): 2235-2240

The purpose of the present study was to investigate the biodegradation kinetics in aerobic and anaerobic soil of the following brominated flame retardants: 2,4,4′-tribromodiphenyl ether (BDE 28), decabromodiphenyl ether (BDE 209), tetrabromobisphenol A (TBBPA), 1,2-dibromo-4-(1,2-dibromoethyl)cyclohexane (TBECH), 2,4,6-tribromophenol (246BrPh), and hexabromobenzene (HxBrBz). For comparison, the biodegradation of the chlorinated compounds 2,4,4′-trichlorodiphenyl ether (CDE 28), 2,4,6-trichlorophenol (246ClPh), hexachlorobenzene (HxClBz), and 2,2′,4,4′,5,5′-hexachlorobiphenyl (PCB 153) was also assessed. In aerobic soil, BDE 209 showed no significant degradation during the test period, but concentrations of the other BFRs declined, with half-lives decreasing in the following order: BDE 28 > TBBPA > TBECH > HxBrBz > 246BrPh. Declines in almost the same order were observed in anaerobic soil: BDE 28, BDE 209 > TBBPA > HxBrBz > TBECH >246BrPh.

Intra- and extrapolated half-lives in soil of tested brominated flame retardants ranged from 7 days for 2,4,6-tribromorophenol to >400 days for decabromodiphenyl ether.

Keywords: Brominated flame retardants; Persistence; Soil; Temperature; Sludge

Heidemarie Schaar[a], Manfred Clara[b], Oliver Gans[b] and Norbert Kreuzinger[a]. ([a]Institute of Water Quality, Resources and Waste Management, Vienna University of Technology, Karlsplatz 13/226, 1040 Vienna, Austria, [b] Umweltbundesamt, Spittelauer Lande 5, 1090 Vienna, Austria). Micropollutant removal during biological wastewater treatment and a subsequent ozonation step. Environmental Pollution, Volume 158(5) (2010): 1399-1404

The design criteria for wastewater treatment plants (WWTP) and the sludge retention time, respectively, have a significant impact on micropollutant removal. The upgrade of an Austrian municipal WWTP to nitrogen removal (best available technology, BAT) resulted in increased elimination of most of the analyzed micropollutants. Substances, such as bisphenol-A, 17α-ethinylestradiol and the antibiotics erythromycin and roxithromycin were only removed after the upgrade of the WWTP. Nevertheless, the BAT was not sufficient to completely eliminate these compounds. Thus, a pilot scale ozonation plant was installed for additional treatment of the effluent. The application of 0.6 g O_3 g DOC^{-1} increased the removal of most of the micropollutants, especially for compounds that were not degraded in the previous biological process, as for example carbamazepine and diclofenac. These results indicated that the ozonation of WWTP effluent is a promising technology to further decrease emissions of micropollutants from the treatment process.

SRT is an important criterion for micropollutant removal in wastewater treatment and the application of ozone is suitable for further removal of micropollutants.

Keywords: Micropollutants; Wastewater treatment; Advanced treatment; Ozonation pilot plant

Kristen N. Savage [1,2] , Lee R. Krumholz [2] , Lisa M. Gieg [2] , Victoria A. Parisi [2] , Joseph M. Suflita [2] , Jon Allen [3] , R. Paul Philp [3] & Mostafa S. Elshahed [1] ([1]Department of Microbiology and Molecular Genetics, Oklahoma State University, Stillwater, OK, USA ; [2] Department of Botany and Microbiology and Institute for Energy and the Environment, University of Oklahoma, Norman, OK, USA ; and [3] School of Geology and Geophysics, University of Oklahoma, Norman, OK, USA . Correspondence: Mostafa S. Elshahed, Department of Microbiology and Molecular Genetics, Oklahoma State University, 1110 S Innovation way, Stillwater, OK 74074, USA. Tel.: +1 405 744 3005; fax: +1 405 744 1112; e-mail: mostafa@okstate.edu). Biodegradation of low-molecular-weight alkanes under mesophilic, sulfate-reducing conditions: metabolic intermediates and community patterns. FEMS Microbiology Ecology, Volume 72 (3) (2010_: 485 – 495

We evaluated the ability of the native microbiota in a low-temperature, sulfidic natural hydrocarbon seep (Zodletone) to metabolize short-chain hydrocarbons. *n*-Propane and *n*-pentane were metabolized under sulfate-reducing conditions in initial enrichments and in sediment-free subcultures. Carbon isotope analysis of residual propane in active enrichments showed that propane became enriched in ^{13}C by 6.7 (±2.0)‰, indicating a biological mechanism for propane loss. The detection of *n*-propylsuccinic and isopropylsuccinic acids in active propane-degrading enrichments provided evidence for anaerobic biodegradation via a fumarate addition pathway. A eubacterial 16S rRNA gene survey of sediment-free enrichments showed that the majority of the sequenced clones were phylogenetically affiliated within the *Deltaproteobacteria*. Such sequences were most closely affiliated with clones retrieved from hydrocarbon-impacted marine ecosystems, volatile fatty acid metabolizers, hydrogen users, and with a novel Deltaproteobacterial lineage. Other cloned sequences were affiliated with the *Firmicutes* and *Chloroflexi* phyla. The sequenced clones were only distantly (<95%) related to other reported

low-molecular-weight alkane-degrading sulfate-reducing populations. This work documents the potential for anaerobic short-chain *n*-alkane metabolism for the first time in a terrestrial environment, provides evidence for a fumarate addition mechanism for *n*-propane activation under these conditions, and reveals microbial community members present in such enrichments.

Keywords: low-molecular-weight *n*-alkanes • anaerobic biodegradation • sulfate reduction • microbial community analysis

Jason T. DeJong[a], Brina M. Mortensen[b], Brian C. Martinez[b] and Douglas C. Nelson[c]. ([a]Department of Civil and Environmental Engineering, University of California, Davis, CA 95616, 530 754 8995, United States, [b] Department of Civil and Environmental Engineering, University of California, Davis, CA 95616, United States, [c] Department of Microbiology, University of California, Davis, CA 95616, 530 752 6183, United States). **Bio-mediated soil improvement. Ecological Engineering, Volume 36(2) (2010): 197-210**

New, exciting opportunities for utilizing biological processes to modify the engineering properties of the subsurface (e.g. strength, stiffness, permeability) have recently emerged. Enabled by interdisciplinary research at the confluence of microbiology, geochemistry, and civil engineering, this new field has the potential to meet society's ever-expanding needs for innovative treatment processes that improve soil supporting new and existing infrastructure. This paper first presents an overview of bio-mediated improvement systems, identifying the primary components and interplay between different disciplines. Geometric compatibility between soil and microbes that restricts the utility of different systems is identified. Focus is then narrowed to a specific system, namely bio-mediated calcite precipitation of sands. Following an overview of the process, alternative biological processes for inducing calcite precipitation are identified and various microscopy techniques are used to assess how the pore space volume is altered by calcite precipitation, the calcite precipitation is distributed spatially within the pore space, and the precipitated calcite degrades during loading. Non-destructive geophysical process monitoring techniques are described and their utility explored. Next, the extent to which various soil engineering properties is identified through experimental examples. Potential advantages and envisioned applications of bio-mediated soil improvement are identified. Finally, the primary challenges that lie ahead, namely optimization and upscaling of the processes and the education/training of researchers/practitioners are briefly discussed.

Keywords: Bio-mediated; Bio-soil; Soil improvement; Ground improvement; Geotechnical engineering; Calcite precipitation; Microbes; Microbial induced calcite precipitation

Hong-Yan Zeng[1], He Jiang[1], Kui Xia[1], Ya-Ju Wang[1] and Yan Huang[1]. ([1]Institute of Biotechnology, College of Chemical Engineering, University of Xiangtan, Xiangtan, 411105, Hunan, China). **Characterization of phenol degradation by high-efficiency binary mixed culture. Environmental Science and Pollution Research, Volume 17(5) (2010): 1035-1044**

Background, aim, and scope

Two new high phenol-degrading strains, *Micrococcus* sp. and *Alcaligenes faecalis* JH 1013, were isolated. The two isolates could grow aerobically in mineral salts medium containing

phenol as a sole carbon source at concentration of 3,000 mg L^{-1}. It was found that the binary mixed culture of the two isolates possessed good potential for phenol removal.

Material and methods

Phenol biodegradation using the binary mixed culture of the two isolates was studied. The optimal conditions were determined to be temperature 32Â°C, pH 7.0, inoculum size 10.0%, and agitation rate 150 rpm in the synthetic wastewater. In addition, the kinetics of the cell growth and phenol degradation by the binary mixed culture were also investigated using Haldane model over a wide range of initial phenol concentrations from 20 to 2,400 mg L^{-1}.

Results

The experimental data indicated that the binary mixed culture had pretty high phenol degradation potential, which could thoroughly degrade the phenol in the synthetic wastewater containing phenol 2,400 mg L^{-1} within 72 h under aerobic condition. Under the optimal conditions, the phenol concentration was reduced speedily from 1,000 to below 0.28 mg L^{-1} in the presence of the binary mixed culture, and the phenol degradation rate reached 99.97% after 16 h. It was well below the standard value 0.28 mg L^{-1} as described by Chinese Environmental Protection Agency. It was clear that the Haldane kinetic model adequately described the dynamic behavior of phenol degradation by the binary mixed culture with kinetic constants of q_{max}=0.45 h^{-1}, K_{sq}=64.28 mg L^{-1}, and K_{iq}=992.79 mg L^{-1}. The phenol concentration to avoid substrate inhibition had been inferred theoretically to be 252.62 mg L^{-1}.

Conclusions

Phenol, as the only carbon source, could be degraded by the binary mixed culture at high initial phenol concentrations. Phenol exhibited inhibitory behavior, and the growth kinetics of the binary mixed culture could be correlated well by the simple Haldane's inhibitory model. The kinetics parameters were invariably required for the design and simulation of batch and continuous bioreactor treating phenolic wastewaters.

Keywords: Biodegradation - Phenol - Kinetic - *Micrococcus* sp. - *Alcaligenes faecalis*

Celine Justino[1], Ana Gabriela Marques[1], KÃ¡tia Reis Duarte[1], Armando Costa Duarte[2], Ruth Pereira[3], Teresa Rocha-Santos[1] and Ana Cristina Freitas. ([1]ISEIT/Viseu, Instituto Piaget, Estrada do Alto do Gaio, Galifonge, 3515-776, Lordosa, Viseu, Portugal, [2]CESAM (Centro de Estudos do Ambiente e do Mar) & Departamento de QuÃmica, Universidade de Aveiro, Campus de Santiago, 3810-193 Aveiro, Portugal, [3]CESAM (Centro de Estudos do Ambiente e do Mar) & Departamento de Biologia, Universidade de Aveiro, Campus de Santiago, 3810-193 Aveiro, Portugal). **Degradation of phenols in olive oil mill wastewater by biological, enzymatic, and photo-Fenton oxidation. Environmental Science and Pollution Research, Volume 17(3) (2010): 650-656**

Background, aim, and scope

Olive oil mill wastewater (OOMW) environmental impacts minimization have been attempted by developing more effective processes, but no chemical or biological treatments were found to be totally effective to mitigate their impact on receiving systems. This work is the first that reports simultaneously the efficiency of three different approaches: biological treatment by two

fungal species (*Trametes versicolor* or *Pleurotus sajor caju*), enzymatic treatment by laccase, and chemical treatment by photo-Fenton oxidation on phenols removal.

Materials and methods

Those treatments were performed on OOMW with or without phenol supplement (*p*-coumaric, vanillin, guaiacol, vanillic acid, or tyrosol). OOMW samples resulted from treatments were extracted for phenols using liquid–liquid extraction and analyzed by gas chromatography coupled to mass spectrometry.

Results

Treatment with *T. versicolor* or *P. sajor caju* were able to remove between 22% and 74% and between 8% and 76% of phenols, respectively. Treatment by laccase was able to reduce 4% to 70% of phenols whereas treatment by photo-Fenton oxidation was responsible for 100% phenols reduction.

Discussion

Range of phenol degradation was equivalent between *T. versicolor*, *P. sajor caju* and laccase for *p*-coumaric, guaiacol, caffeic acid, and tyrosol in supplemented OOMW, which enhances this enzyme role in the biological treatment promoted by these two species.

Conclusions

Phenols were removed more efficiently by photo-Fenton treatment than by biological or enzymatic treatments.

Recommendations and perspectives

Use of fungi, laccase, or photo-Fenton presents great potential for removing phenols from OOMW. This should be further assessed by increasing the application scale and the reactor configurations effect on the performance, besides a toxicity evaluation of treated wastewater in comparison to raw wastewater.

Keywords: Olive oil mill wastewater - Phenols - Photo-Fenton oxidation - Laccase - *Pleurotus sajor caju* - *Trametes versicolor*

Huda Mahmoud[1], Redha Al-Hasan[1], Majida Khanafer[1] and Samir Radwan[1] . ([1]Faculty of Science, Department of Biological Sciences, Kuwait University, P. O. Box 5969, 13060 Safat, Kuwait). A microbiological study of the self-cleaning potential of oily Arabian Gulf coasts. Environmental Science and Pollution Research, Volume 17(2) (2010): 383-391

Background, aim, and scope Due to the active production and transport of crude oil in the Arabian Gulf region, the Arabian Gulf coasts are routinely polluted with oil. Therefore, such coasts have been subject of studies aiming at assessing the roles of indigenous microbial consortia in cleaning these environments. In the present study, epilithic microbial communities along Kuwait coasts were studied for their oil degradation potential.

Materials and methods Gravel particles coated with deep green biofilms were collected from four coastal sites in autumn, winter, and spring. Phototrophs in these consortia were determined in terms of their chlorophyll a contents and identified by their morphological characteristics. Total bacteria were counted microscopically and cultivable bacteria by the dilution plating method on nutrient agar as well as on inorganic medium containing oil as a sole source of carbon and energy. The bacterial community structures were also characterized and compared by denaturing gradient gel electrophoresis (DGGE).

Results Epilithic biomass samples from the four sites in the three seasons were rich in diatoms and picocyanobacteria as well as total bacteria. Direct counting gave bacterial numbers per square centimeter gravel surface of 2 to 6 Ã— 10^7 cells depending on the sampling site and season. Cultivable bacterial numbers on nutrient agar and crude oil as a sole source of carbon were 3 Ã— 10^3 to 8 Ã—10^4 and 1 Ã— 10^3 to 7 Ã— 10^3 cells/cm^2 gravel surface, respectively. The DGGE profiles of epilithon biomass samples revealed major 16S rDNA bands that matched bands of pure oil-utilizing bacterial isolates.

Discussion The microbial communities showed a degree of consistency in all sites and seasons.

Conclusions The microbial consortia coating gravel particles are potentially suitable tools for self-cleaning of oily Gulf coasts. They are rich in oil-utilizing bacteria whose activities are probably enhanced by oxygen produced by the phototrophic partners in the consortia.

Recommendations and perspectives The combination of conventional microbiological analysis with molecular approaches gives an enhanced idea about natural microbial communities especially those with environmental application potential.

Keywords: Arabian Gulf - Bioremediation - DGGE - Epilithon consortia - Oil utilization - Phototrophic microorganisms

Brian A. Pellerin[a,*], Peter J. Hernes[b], JohnFranco Saraceno[a], Robert G. M. Spencer[b] and Brian A. Bergamaschi[a] . ([a] U.S. Geological Survey, 6000 J Street, Placer Hall, Sacramento, CA 95819, [b] Dep. Of Land, Air and Water Resources, Univ. of California, Davis, CA 95616. Assigned to Associate Editor Lakhwinder Hundal. Corresponding author (bpeller@usgs.gov). © ASA, CSSA, SSSA 5585 Guilford Rd., Madison, WI 53711 USA). Microbial Degradation of Plant Leachate Alters Lignin Phenols and Trihalomethane Precursors. Journal of Environ Qual 39 (2010): 946-954

Received for publication December 9, 2009. Although the importance of vascular plant–derived dissolved organic carbon (DOC) in freshwater systems has been studied, the role of leached DOC as precursors of disinfection byproducts (DBPs) during drinking water treatment is not well known. Here we measured the propensity of leachates from four crops and four aquatic macrophytes to form trihalomethanes (THMs)—a regulated class of DBPs—before and after 21 d of microbial degradation. We also measured lignin phenol content and specific UV absorbance ($SUVA_{254}$) to test the assumption that aromatic compounds from vascular plants are resistant to microbial degradation and readily form DBPs. Leaching solubilized 9 to 26% of total plant carbon, which formed 1.93 to 6.72 mmol THM mol C^{-1}. However, leachate DOC concentrations decreased by 85 to 92% over the 21-d incubation, with a concomitant decrease of 67 to 92% in total THM formation potential. Carbon-normalized THM yields in the residual DOC pool increased by 2.5 times on average, consistent with the preferential uptake of nonprecursor

material. Lignin phenol concentrations decreased by 64 to 96% over 21 d, but a lack of correlation between lignin content and THM yields or $SUVA_{254}$ suggested that lignin-derived compounds are not the source of increased THM precursor yields in the residual DOC pool. Our results indicate that microbial carbon utilization alters THM precursors in ecosystems with direct plant leaching, but more work is needed to identify the specific dissolved organic matter components with a greater propensity to form DBPs and affect watershed management, drinking water quality, and human health.

Abbreviations: DBP, disinfection byproducts • DOC, dissolved organic carbon • DOM, dissolved organic matter • STHMFP, specific trihalomethane formation potential • THM, trihalomethane • THMFP, trihalomethane formation potential

Ren-bang ZHAO[a, b], Hua-ying BAO[a] and Yuan-xia LIU[a]. ([a] College of Chemistry, Beijing Normal University, Beijing 100875, P.R.China, [b] Faculty of Food Science and Technology, Agricultural University of Hebei, Baoding 071001, P.R.China). **Isolation and Characterization of *Penicillium oxalicum* ZHJ6 for Biodegradation of Methamidophos. Agricultural Sciences in China, Volume 9(5) (2010): 695-703**

One methamidophos-degrading fungus strain, named as ZHJ6, was isolated from the soils contaminated with methamidophos. It was identified as *Penicillium oxalicum* based on its morphological characteristics and ITS rDNA gene sequence analysis. The effects of carbon source, nitrogen source and the concentration of methamidophos, temperature and pH on the degradation were investigated. The results showed that the strain could use glucose as carbon source and the methamidophos as sole nitrogen source. The degradation ratio of methamidophos, when the initial concentration was 1.0×10^{-3} mg mL^{-1}, could reach above 99.9% in 12 incubation days. The strain could use ethanol, glucose, fructose, sucrose, lactose, starch, and dextrin as its carbon and energy source to degrade the methamidophos. The favorable degrading condition of the strain ZHJ6 was in a mineral salt medium at pH 5.0 and 25°C with 1% glucose, and further studies showed that the strain could degrade folimat, phoxim and glyphosate with glucose as carbon source, but could not degrade chlorpyrifos, phosdrin, trichlorphon, and dichlorvos.

Key words: fungus; biodegradation; methamidophos; *Penicillium oxalicum*; pesticide; remediation

Cun-zheng ZHANG[a], Xin-ming ZHANG[d], Zi-hua TIAN[b], Dan-jun HE[c] and Xian-jin LIU[a]. ([a] Institute of Food Safety, Jiangsu Academy of Agricultural Sciences/Key Laboratory of Food Safety Monitoring and Management, Ministry of Agriculture/Key Lab of Food Quality and Safety of Jiangsu Province - State Key Laboratory Breeding Base, Nanjing 210014, P.R. China, [b] Nanjing Centre of Inspection and Testing for Agricultural Products, Ministry of Agriculture, Nanjing 210036, P.R.China, [c] Nanjing Exit-Entry Inspection and Quarantine Bureau, Nanjing 210003, P.R.China, [d] Taicang Centre of Inspection and Testing for Agricultural Products, Taicang 215412, P.R.China). **Degradation of Chlorpyrifos and Fipronil in Rice from Farm to Fork and Risk Assessment. Agricultural Sciences in China, Volume 9(5) (2010): 754-763**

Degradation of pesticide residues (chlorpyrifos and fipronil) in rice from farm to fork and risk assessment for human health were studied to reveal the magnitude of risks faced by different populations of interest, so that appropriate measures can be taken to control the risks, and to refine and update the human health risk assessment data while helping to determine the maximum residue level (MRL) value and harvest interval. Different dosages and treatments were used in field trials for the harvest residue test. Residue levels of postharvest-applied chlorpyrifos and fipronil during storage, exposure to sunlight, washing and boiling processes (boiled rice) were investigated for brown rice. The dietary exposure evaluation model (DEEM) was employed to estimate acute and chronic risks faced by different populations of interest. Percent of reference dose (POR) and margin of exposure (MOE) were calculated. A positive correlation between pesticide residues and the dosage and application frequency of pesticide was found in the field trials. Risk quotients indicate that multiple applications and double dosages of chlorpyrifos increase the risks to the entire population and prolong exposures to toxic concentrations. The concentration of pesticide residues decreased as a function of time, after sunlight exposure, storage, washing, and boiling processes. 91.6 and 96.16% degradations were achieved at the end of the experimental period for fipronil and chlorpyrifos, respectively. The boiling process played an important role in the degradation of these pesticides. The result of risk assessment to human health showed that harvest residues of chlorpyrifos in rice and acute dietary risks of chlopyrifos were of concern. The acute dietary (food only) risk estimated for chlorpyrifos as percent of acute population adjusted dose (aPAD) was frequently over 100%. The risk faced by boys under the age of 14 was higher than that for girls of the same age. For the subpopulation above age 14, the risk reversed. The chronic dietary risk from food alone showed that dietary exposures with fipronil were below the level of concern for the entire population, including children. The risk faced by rural residents was more serious than that for urbanite residents with the most sensitive populations being children and male residents who faced higher acute dietary risk than the other subpopulation groups. The harvest interval was found to be the critical measure to mitigate risk for all populations for safe rice eating. All risk levels decreased to acceptable levels when the harvest interval was extended to 14 d. To address these risks, a number of measures including reduced application rates (should not be doubled at single application), increased retreatment intervals (longer than 7 d) and extended interval of harvest (at least 14 d) will be needed. The MRL for fipronil in rice is recommended to be 0.01 mg kg^{-1} in accordance with Codex (ref).

Key words: rice; risk assessment; pesticide residue; chlorpyrifos; fipronil

J.W. Hong, J.Y. Park and G.M. Gadd. (Division of Molecular Microbiology, College of Life Sciences, University of Dundee, Dundee, UK, Correspondence to Geoffrey Michael Gadd, Division of Molecular Microbiology, College of Life Sciences, University of Dundee, Dundee DD1 5EH, UK. E-mail: g.m.gadd@dundee.ac.uk). **Pyrene degradation and copper and zinc uptake by Fusarium solani and Hypocrea lixii isolated from petrol station soil. Journal of Applied Microbiology,** Volume 108(6) (2010): 2030 - 2040

Aims: This study aimed to isolate and identify potential polycyclic aromatic hydrocarbon (PAH)-degrading and/or metal-tolerant fungi from PAH-contaminated and metal-contaminated soils.

Methods and Results: Pyrene-degrading fungi were isolated from contaminated soil and tested for metal (Cu, Zn and Pb) compound solubilization and metal accumulation. Three strains of Fusarium solani and one of Hypocrea lixii were able to degrade more than 60% of initial supplied pyrene (100 mg l^{-1}) after 2 weeks. The isolates were grown on toxic metal (Cu, Pb and

Zn)-containing media: all isolates accumulated Cu in their mycelia to values ranging from c. 5·9 to 10·4 mmol per kg dry weight biomass. The isolates were also able to accumulate Zn (c. 3·7–7·2 mmol per kg dry weight biomass) from zinc phosphate-amended media. None of the isolates accumulated Pb.

Conclusions: These fungal isolates appear to show promise for use in bioremediation of pyrene or related xenobiotics and removal of copper and zinc from wastes contaminated singly or in combination with these substances.

Significance and Impact of the Study: Microbial responses to mixed organic and inorganic pollution are seldom considered: this research highlights the abilities of certain fungal strains to interact with both xenobiotics and toxic metals and is relevant to other studies on natural attenuation and bioremediation of polluted sites.

Youngmin Lee[a] and Woojin Lee[a]. ([a] Department of Civil and Environmental Engineering, Korea Advanced Institute of Science and Technology, 373-1 Guseong-Dong, Yuseong-Gu, Daejon 305-701, Republic of Korea). Degradation of trichloroethylene by Fe(II) chelated with cross-linked chitosan in a modified Fenton reaction. Journal of Hazardous Materials, Volume 178(1-3) (2010): 187-193

Degradation of trichloroethylene (TCE) by a modified Fenton reaction was investigated in aqueous solution. Fenton reaction can be significantly enhanced in the presence of Fe(II) chelated by cross-linked chitosan (CS) with glutaraldehyde (GLA) at neutral pH. A remarkable oxidative degradation of TCE (1.838 h^{-1}) was observed in the modified Fenton system with Fe(II)-CS/GLA (10 mM and 2 g L^{-1}, respectively) and H_2O_2 (318 mM), while no significant degradation (0.005 h^{-1}) was observed in the classic Fenton reaction system with Fe(II) (10 mM) and H_2O_2 (318 mM) at pH 7 in 5 h. The kinetic rate constants for the degradation of TCE in the modified Fenton system was dependent on the initial suspension pH, Fe(II) loading, CS/GLA dosage, and concentration of H_2O_2. We observed the formation of surface Fe(II)-CS/GLA complex using microscopic analyses and identified Fe oxidation (Fe(II) to Fe(III)) coupled with H_2O_2 reduction on the Fe(II)-CS/GLA surfaces during the modified Fenton reaction.

Keywords: Modified Fenton reaction; Cross-linked chitosan; Fe(II) chelation; Neutral pH; TCE

Antonella Anastasi[a], Valeria Prigione[a] and Giovanna Cristina Varese[a]. ([a] Department of Plant Biology, University of Turin, Viale Mattioli 25, 10125 Turin, Italy). Industrial dye degradation and detoxification by basidiomycetes belonging to different eco-physiological groups. Journal of Hazardous Materials, Volume 177(1-3) (2010): 260-267

Twenty-five basidiomycetes belonging to 17 species and ascribable to different eco-physiological groups were screened for their ability to decolorize 9 commercially important industrial dyes comprising a variety of anthraquinonic, azoic and phtalocyanin chromophores. The influence of the culture medium, particularly its C:N ratio, on decolourisation capacity was considered on solid substrate. Three strains of *Bjerkandera adusta* performed the highest decolourisation yields being able to degrade all dyes on all media and to produce a wide spectrum of oxidative enzyme activities. Hence, *B. adusta* strains were selected for further experiments in liquid cultures together with other 6 fungi that resulted effective in the

decolourisation of the largest number of molecules in the broadest spectrum of cultural conditions. Particularly *B. adusta* MUT 3060 was found very effective (decolourisation percentage over 90%) in the treatment of simulated effluents composed of single and mixed dyes at high concentration (1000 mg/l). Peroxidase activity dependent (up to 362 U/l) and independent from manganese (up to 57 U/l) were detected during the decolourisation process. The *Lemna minor* toxicity test showed a significant reduction of toxicity after the fungal treatment indicating that decolourisation corresponded to an actual detoxification of the wastewater.

Keywords: Biodegradation; Basidiomycetes; Ecotoxicity; Industrial dyes; Peroxidases

C.-H. Chen[a], C.-F. Chang[a] and S.-M. Liu[a]. ([a] Institute of Marine Biology, National Taiwan Ocean University, 2-Peining Rd., Keelung, Taiwan). Partial degradation mechanisms of malachite green and methyl violet B by *Shewanella decolorationis* NTOU1 under anaerobic conditions. Journal of Hazardous Materials, Volume 177(1-3) (2010): 281-289

This work demonstrated that *Shewanella decolorationis* NTOU1 decolorized 200 mg l^{-1} of crystal violet, malachite green, or methyl violet B within 2–11 h under anaerobic conditions at 35 °C. The initial color removal rate of malachite green was highest, while that of methyl violet was lowest. GC/MS analyses of the intermediate compounds produced during and after decolorization of malachite green and methyl violet B suggested that biodegradation of these dyes involved reduction to leuco form, N-demethylation, and reductive splitting of the triphenyl rings. The number of N-methylated groups of these dyes might have influenced decolorization rates and the reductive splitting of the triphenyl rings of these dyes. Cytotoxicity and antimicrobial test data showed that malachite green and methyl violet B solution (100 mg l^{-1}) were toxic. Toxicity of the dyes decreased after their decolorization, but further incubation resulted in increased toxicity.

Keywords: Decolorization; Malachite green; Methyl violet B; Partial degradation mechanisms; *Shewanella decolorationis* NTOU1

Sardar Alam Cheema[a, b], Muhammad Imran Khan[a, b], Chaofeng Shen[a], Xianjin Tang[a], Muhammad Farooq[b], Lei Chen[a], Congkai Zhang[a] and Yingxu Chen[a]. ([a]Institute of Environmental Science and Technology, Zhejiang University, 268 Kaixuan Road, Hangzhou 310029, Zhejiang, PR China, [b] Department of Agronomy, University of Agriculture, Faisalabad 38040, Pakistan). Degradation of phenanthrene and pyrene in spiked soils by single and combined plants cultivation. Journal of Hazardous Materials, Volume 177(1-3) (2010): 384-389

The present study was conducted to investigate the capability of four plant species (tall fescue, ryegrass, alfalfa, and rape seed) grown alone and in combination to the degradation of phenanthrene and pyrene (polycyclic aromatic hydrocarbons, PAHs) in spiked soil. After 65 days of plant growth, plant biomass, dehydrogenase activity, water-soluble phenolic (WSP) compounds, plant uptake and accumulation and residual concentrations of phenanthrene and pyrene were determined. Our results showed that presence of vegetation significantly enhanced the dissipation of phenanthrene and pyrene from contaminated soils. Higher degradation rates of PAHs were observed in the combined plant cultivation (98.3–99.2% phenanthrene and 88.1–95.7% pyrene) compared to the single plant cultivation (97.0–98.0% phenanthrene and 79.8–86.0% pyrene). Contribution of direct plant uptake and accumulation of phenanthrene and

pyrene was very low compared to the plant enhanced dissipation. By contrast, plant-promoted biodegradation was the predominant contribution to the remediation enhancement. The correlation analysis indicates a negative relation between biological activities (dehydrogenase activity and WSP compounds) and residual concentrations of phenanthrene and pyrene in planted soils. Our results suggest that phytoremediation could be a feasible choice for PAHs contaminated soil. Moreover, the combined plant cultivation has potential to enhance the process.

Keywords: PAHs; Phenanthrene; Pyrene; Phytoremediation; Combined plant cultivation

Congying Wang[a, b], Fang Wang[a], Tao Wang[a], Yongrong Bian[a], Xinglun Yang[a] and Xin Jiang[a]. ([a] State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing 210008, China, [b] School of Environmental Science and Resources, Shanxi University, Taiyuan 030006, China). **PAHs biodegradation potential of indigenous consortia from agricultural soil and contaminated soil in two-liquid-phase bioreactor (TLPB). Journal of Hazardous Materials, Volume 176(1-3) (2010): 41-47**

Estimation of PAHs degradation potential of indigenous consortia is essential for remediation of polluted soils. In this study, the biodegradation of a mixture of 11 PAHs was compared using a long-term PAH-contaminated soil (CS) and an unpolluted agricultural soil (AS) as inocula in a two-liquid-phase bioreactor (TLPB). In the TLPB, silicone oil was used as the organic phase to increase the PAHs bioavailability. The microbial numbers were also determined during the biodegradation. The results demonstrated that naphthalene, fluorene, phenanthrene, anthracene, fluoranthene and pyrene could be completely biodegraded in both soils within 4–50 days. With the exception of dibenzo(a,h)anthrancene, the other PAHs including benzo(a)anthracene, benzo(a)pyrene, benzo(b)fluoranthene and benzo(k)fluoranthene were degraded to different extents in both soils at the end of 170 days. Complete biodegradation of benzo(a)anthracene and benzo(b)fluoranthene only occurred in CS. During the process, microbial growth was highly correlated to the biodegradation of PAHs. Sequential utilization of PAHs showed a competitive-inhibition in the multi-substrate system. The half-life times of PAHs obtained here were much shorter than those reported previously in soils, indicating that indigenous microbes in both soils had high PAHs degradation potential, facilitated by TLPB.

Keywords: PAHs biodegradation; Indigenous consortia; Microbial activity; Sequential utilization; Two-liquid-phase bioreactor

Andrea Dario[a], Marc Schroeder[a], Gibson S. Nyanhongo[a], Gregor Englmayer[b] and Georg M. Guebitz[a]. ([a] Institute of Environmental Biotechnology, Graz University of Technology, Petersgasse 12, A-8010 Graz, Austria, [b] Austin Powder GmbH, St. Lambrecht, Austria). **Development of a biodegradable ethylene glycol dinitrate-based explosive. Journal of Hazardous Materials, Volume 176(1-3) (2010): 125-130**

Bacteria capable of degrading ethylene glycol dinitrate (EGDN) were isolated under aerobic and, carbon and nitrogen-limiting conditions from EGDN contaminated soil and rivers. EGDN degradation was monitored using HPLC and UV–Vis spectrometer. Among the isolated strains *Bacillus subtilis* GN was the best, completely degrading 6.6 mM EGDN with the concomitant

release of nitrite and EGMN within 72 h. Furthermore, the level of toxicity of EGDN as measured by the bioluminescent bacteria, *Vibrio fischeri* was reduced by 80% when 100% of the 6.6 mM EGDN was degraded. An environmentally friendly "biodegradable explosives", was achieved by adsorbing *B. subtilis* GN spores onto the wood flour, an ingredient of the explosive. The incorporation of *B. subtilis* GN spores into the explosive formulation did not affect the quality of the explosive as confirmed by the almost unchanged detonation velocity (3410 ms^{-1} compared to 3500 ms^{-1} of the control), autoignition temperature, Abel test, shock and friction sensitivity test. It was also possible to achieve rapid degradation of the residues after detonation upon exposure to air and moisture.

Keywords: Ethylene glycol dinitrate; Biodegradation; Toxicity test

Lin Qiao[a, b] and Jian-long Wang[a]. ([a] Laboratory of Environmental Technology, INET, Tsinghua University, Beijing 100084, PR China, [b] Beijing Wei Ming Kai Tuo Agriculture Biotechnology Co., Ltd., Beijing 100085, PR China). **Microbial degradation of pyridine by *Paracoccus* sp. isolated from contaminated soil. Journal of Hazardous Materials, Volume 176(1-3) (2010): 220-225**

A pyridine-degrading strain was isolated from the contaminated soil near the pesticide plant, identified as *Paracoccus* sp., and designated as strain KT-5, on the basis of its partial 16S rRNA gene sequence analysis. The effect of different co-substrates including glucose, ammonium chloride and trace elements on biodegradation of pyridine by *Paracoccus* sp. KT-5 was investigated. The results showed that when the initial concentration of pyridine was about 900 mg L^{-1}, 100 mg L^{-1} of glucose increased the growth of strain KT-5 and the removal of pyridine, but did not affect the release of nitrogen in the pyridine ring as ammonia. In addition, strain KT-5 was able to utilize 100 mg L^{-1} of glucose and 900 mg L^{-1} of pyridine simultaneously as the carbon source. 100 mg L^{-1} of ammonium chloride inhibited the growth of strain KT-5 in 900 mg L^{-1} of pyridine, and also slightly decreased the removal of pyridine, but did not affect the release of nitrogen in the pyridine ring as ammonia. However, lacking of trace elements not only inhibited the growth of strain KT-5 in 900 mg L^{-1} of pyridine, but also decreased the removal of pyridine, while it did not affect the release of nitrogen in the pyridine ring as ammonia.

Keywords: Pyridine; Biodegradation; Glucose; Nitrogen source; Trace elements

Xueling Wu[a], Renxing Liang[a], Qinyun Dai[a], Decai Jin[a], Yangyang Wang[a] and Weiliang Chao[b]. ([a]Department of Bioengineering, School of Minerals Processing and Bioengineering, Central South University, Changsha 410083, China, [b] Department of Microbiology, Soochow University, Taipei, Taiwan). **Complete degradation of di-*n*-octyl phthalate by biochemical cooperation between *Gordonia* sp. strain JDC-2 and *Arthrobacter* sp. strain JDC-32 isolated from activated sludge. Journal of Hazardous Materials, Volume 176(1-3) (2010): 262-268**

Two bacterial strains were isolated from activated sludge using mixtures of phthalic acid esters (PAEs) as the sole source of carbon and energy. One of the isolates was identified as *Gordonia* sp. strain JDC-2 and the other as *Arthrobacter* sp. strain JDC-32, mainly through 16S rRNA gene sequence analysis. *Gordonia* sp. strain JDC-2 rapidly degraded di-*n*-octyl phthalate (DOP) into phthalic acid (PA), which accumulated in the culture medium. *Arthrobacter* sp. strain JDC-32 degraded PA but not DOP. The co-culture of *Gordonia* sp. strain JDC-2 and *Arthrobacter* sp.

strain JDC-32 degraded DOP completely by overcoming the degradative limitations of each species alone. The biochemical pathway of DOP degradation by *Gordonia* sp. strain JDC-2 was proposed based on the identified degradation intermediates. The results suggest that DOP is completely degraded by the biochemical cooperation of different microorganisms isolated from activated sludge.

Keywords: Degradation; Di-*n*-octyl phthalate; *Arthrobacter* sp.; *Gordonia* sp.; Biochemical cooperation

Inés Infante[1], Maria A. Morel[1], Martha C. Ubalde[1], Cecilia Martínez-Rosales[1], Silvia Belvisi[2] and Susana Castro-Sowinski[1, 3]. ([1]Unidad de Microbiología Molecular, Instituto Clemente Estable, Av. Italia 3318, Montevideo, Uruguay, [2]Paylana SA, Av. Salto 919, Paysandú, Uruguay, [3]Sección Bioquímica, Facultad de Ciencias, Universidad de la República, Iguá 4225, Montevideo, Uruguay). **Wool-degrading *Bacillus* isolates: extracellular protease production for microbial processing of fabrics. World Journal of Microbiology and Biotechnology, Volume 26(6) (2010): 1047-1052**

Wool is a natural animal fiber commonly used in fabrics, but requires physical and chemical processing treatment for such applications. With the aim of developing new woollen textile products using environmentally friendly treatments, proteolytic bacteria were isolated from raw wool samples of *Merino* sheep and screened for wool-degrading activity. Two isolates were identified as *Bacillus megaterium* L4 and *Bacillus thuringiensis* L11 by 16S rRNA gene sequence analysis. Both isolates grew on a minimal medium using wool-fiber or wool-fabric as sole carbon and nitrogen sources. Bacterial growth was correlated with extracellular protease activity, and maximal protease production was in early stationary phase. The exoprotease produced by L11 was found to be a thermo-tolerant metalloprotease stabilized by calcium or magnesium, and had optimum activity at pH 7.0 and temperature at 40°C. During bacterial growth the wool-fiber lost weight, but it did not show changes in diameter. When wool-fabric was used instead of wool-fiber weight loss and non-shrinking was found. These are encouraging results for textile processing that should be useful for development of new textile products by direct microbial processing. A potential alternative that could be suggested from our study would be to treat wool with wool-degrading microorganisms in order to develop environmentally friendly processes.

Keywords: *Bacillus* - Protease – Wool

Wen-Teish Chang[1], Ming-Lun Chen[1] and San-Lang Wang[2]. ([1]Department of Food Science, National Penghu University, No. 300, Liu-Ho Rd., Makung City, Penghu Hsien, Taiwan, [2]Graduate Institute of Life Sciences, Tamkang University, Taipei, 251, Taiwan). **An antifungal chitinase produced by *Bacillus subtilis* using chitin waste as a carbon source. World Journal of Microbiology and Biotechnology, Volume 26(5) (2010): 945-950**

The production of inexpensive chitinolytic enzymes is an element in the utilization of shellfish-processing waste. In this study, shrimp and crab shell powder, prepared by treating shrimp- and crab-processing waste by boiling and crushing, was used as a substrate for the isolation of an antifungal chitinase-producing microorganism. *Bacillus subtilis* NPU 001, a strain isolated from soil samples, excreted a chitinase when cultured in a medium containing 2% (w/v) shrimp and

crab shell powder as the major carbon source. The chitinase, which was purified by sequential chromatography, had a Mw of 31 kDa and a pI of 5.4. The purified chitinase (2 mg ml^{-1}) inhibited hyphal extension of the fungus *Fusarium oxysporum*. Compared with other known bacterial chitinases, the unique characteristics of NPU 001 chitinase include antifungal activity against plant-pathogenic fungi and the production of chitotriose as the major enzymatic hydrolysate from colloidal chitin.

Keywords: *Bacillus subtilis* - Chitinase - *Fusarium* - Shrimp and crab shell

Ram Chandra[1], Sangeeta Yadav[1] and Ram Naresh Bharagava[1]. ([1]Environmental Microbiology Section, Indian Institute of Toxicology Research (CSIR), Post Office Box No. 80, M. G. Marg, Lucknow, Uttar Pradesh, 226 001, India). Biodegradation of pyridine raffinate by two bacterial co-cultures of *Bacillus cereus* (DQ435020) and *Alcaligenes faecalis* (DQ435021). World Journal of Microbiology and Biotechnology, Volume 26(4) (2010): 685-692

This study deals with the optimization of bacterial degradation of pyridine raffinate by previously isolated two aerobic bacteria ITRCEM1 (*Bacillus cereus*) and ITRCEM2 (*Alcaligens faecalis*) with accession number DQ4335020 and DQ435021, respectively. The degradation of pyridine raffinate was studied by axenic and mixed bacterial consortium at different nutritional and environmental conditions after the removal of formaldehyde from pyridine raffinate (FPPR). Results revealed that the optimum degradation of pyridine raffinate was observed by mixed bacterial culture in presence of glucose (1% w/v) and peptone (0.2% w/v) at 20% FPPR, pH 7.0, temperature 30°C and 120 rpm at 168 h incubation period . The HPLC analysis of degraded pyridine raffinate samples has indicated the complete removal of α, β and γ picoline. Further, the GC–MS analysis of FPPR pyridine raffinate has shown the presence of pyrazine acetonitrile (6.74), 1,3-dioxepin (8.68), 2-pyridine carboxaldehyde (11.26), propiolactone (12.06), 2-butanol (13.10), benzenesulfonic acid (16.22) and 1,4-dimethyl pyperadine while phenol (17.64) and 3,4-dimethyl benzaldehyde as metabolic products of FPPR.

Keywords: *Alcaligenes faecalis* - *Bacillus cereus* - Degradation - GC–MS analysis - Pyridine raffinate

Natalia N. Pozdnyakova[1], Svetlana V. Nikiforova[1], Oleg E. Makarov[1], Marina P. Chernyshova[1], Kirill E. Pankin[1] and Olga V. Turkovskaya[1]. ([1]Institute of Biochemistry and Physiology of Plants and Microorganisms, Russian Academy of Sciences, 13 Prospect Entuziastov, 410049 Saratov, Russia). Influence of cultivation conditions on pyrene degradation by the fungus *Pleurotus Ostreatus* D1. World Journal of Microbiology and Biotechnology, Volume 26(2) (2010): 205-211

For the first time the dependence of completeness of pyrene degradation by the white-rot fungus *Pleurotus ostreatus* D1 on cultivation conditions was found. In Kirk's medium about 65.6 ± 0.9% of the initial pyrene was metabolized after 3 weeks, with pyrene-4,5-dihydrodiol accumulating. This process was accompanied by laccase production only. In basidiomycetes rich medium, *P. ostreatus* D1 metabolized up to 89.8 ± 2.3% of pyrene within 3 weeks without pyrene-4,5-dihydrodiol accumulation throughout the time of cultivation. Phenanthrene and phthalic acid were identified as the metabolites produced from pyrene degradation under these conditions. Accumulation of phenanthrene with its subsequent disappearance was observed. One more metabolite probably was the product of phenanthrene degradation. Pyrene metabolism in

basidiomycetes rich medium was accompanied first by laccase and tyrosinase production and later by versatile peroxidase production. The cell-associated activities of laccase, tyrosinase, and versatile peroxidase were found. The data obtained indicate that both enzymes (laccase and versatile peroxidase) are necessary for complete degradation of pyrene. Furthermore, both cell-associated and extracellular laccases can catalyse the first stages of pyrene degradation, and versatile peroxidase can be necessary for oxidation of the resulting metabolites.

Keywords: Biodegradation - Laccase - *Pleurotus ostreatus* D1 - Pyrene - Versatile peroxidase

C. Sivaraman[1], Anasuya Ganguly[1] and Srikanth Mutnuri[1]. ([1]Biological Sciences Group, Birla Institute of Technology and Science-Pilani, Goa Campus, Zuari Nagar, Goa, 403726, India). **Biodegradation of hydrocarbons in the presence of cyclodextrins. World Journal of Microbiology and Biotechnology, Volume 26(2) (2010): 227-232**

Aliphatic hydrocarbons are one of the main components of oil contamination. Bioremediation is considered to be a cost-effective treatment option among the conventional treatment methods with bioavailability being the limitation. Chemical surfactants could be used to increase the bioavailability of the hydrocarbons but they showed marked toxicity and environmental pollution. Cyclodextrins are cyclic oligosaccharides which can alter the solubility of the hydrocarbons by incorporating suitably sized hydrophobic molecules into their hydrophobic cavities. This paper focuses on studying the degradation of hydrocarbons by *Pseudomonas* like species named as Vid1 isolated previously from bilge oil contaminated waters in the presence of cyclodextrins. Among the three cyclodextrins (α, β and γ) tested at different concentrations, 2.5 mM of β-cyclodextrin showed higher amount of biodegradation when *n*-hexadecane was used as a model hydrocarbon compound. The percentage of residual hexadecane remaining in the 2.5 mM β-cyclodextrin supplied medium at 120 h was found to be 15% in comparison with the biotic control which was 43%. In the next experimental setup, degradation of mixture of hydrocarbons (tetradecane, hexadecane and octadecane) by Vid1 (*Pseudomonas* like species) was studied at a concentration of 2.5 mM β-cyclodextrin. The residual percentage of tetradecane, hexadecane and octadecane at 120 h was found to be 32, 43 and 61% in comparison with the biotic control 50, 58 and 67%, respectively. Our studies show that among a mixture of hydrocarbons (tetradecane, hexadecane and octadecane) in the presence of β-cyclodextrin, the highest concentration of hydrocarbon degradation was found in tetradecane, hexadecane and octadecane, respectively.

Keywords: Hexadecane - Cyclodextrin - Bioavailability – Biodegradation

Bao-Yu Tian[1], Qin-Geng Huang[1], Yan Xu[1], Chun-Xiang Wang[1], Rui-Rui Lv[1] and Jian-Zhong Huang[1]. ([1]Engineering Research Center of Industrial Microbiology of Ministry of Education, College of Life Sciences, Fujian Normal University, 350108 Fuzhou, People's Republic of China). **Microbial community structure and diversity in a native forest wood-decomposed hollow-stump ecosystem. World Journal of Microbiology and Biotechnology, Volume 26(2) (2010): 233-240**

The aim of this study is to investigate the microbial community structure and diversity in a wood-decomposed hollow-stump ecosystem. Microbial communities of SD-1, a lateritic soil sample from forest hollow-stump ecosystems in Fuzhou (a southeastern coast city of China),

were characterized by constructing and analyzing rRNA gene clone libraries. Sixty-six phylotypes were identified from 112 bacterial clones, including *Acidobacteria* (71.5%), *Proteobacteria* (24.1%) and *Verrucomicrobia* (0.9%). A total of 40 phylotypes were obtained from 138 fungal clones, including Basidiomycota (42.8%), Ascomycota (36.2%), Zygomycota (13.8%), Chytridiomycota (2.9%) and Fungi incertae sedis (4.3%). The results showed a variety of clones related to the reported lignocellulose-decomposing microorganisms. They included some important bacterial decomposers, such as *Sphingomonas* and *Burkholderia*, and a number of wood-decaying fungi, including *Tricholomataceae*, *Strophariaceae* and *Agaricaceae* of Basidiomycota; *Orbilia*, *Aspergillus*, *Phialocephala*, *Epicoccum* and *Phoma* of Ascomycota and Mucorales of *Zygomycota*. The result indicated that the lignocellulolytic microorganisms worked synergically with a unique community structure to biodegrade lignocellulose in the hollow-stump ecosystem.

Keywords: Forest hollow-stump ecosystem - RFLP - Phylogenetic analysis - Microbial community - Wood decomposition

Monika Dhote[1, 2], Asha Juwarkar[1] Anil Kumar[2], G. S. Kanade[1] and Tapan Chakrabarti[1]. ([1]Environmental Biotechnology Division, National Environmental Engineering Research Institute (NEERI), Nehru Marg, Nagpur, 440020, India, [2]School of Biotechnology, Devi Ahilya University, Khandwa Road, Indore, 452001, India). **Biodegradation of chrysene by the bacterial strains isolated from oily sludge. World Journal of Microbiology and Biotechnology, Volume 26(2) (2010): 329-335**

The biodegradation studies were conducted to test the ability of the bacterial strains (Chry2 and Chry3) isolated from the oily sludge obtained from Gujarat refinery, India, for utilization of chrysene in the liquid medium. Biodegradation of the compound was confirmed using gas chromatography and the percent degradation was calculated to be 15.0 and 17% by Chry2 and Chry3, respectively. The biodegradation results were supported by increase in viable cell count and dry biomass, in the presence of chrysene as the sole carbon source. Both the cultures produced biosurfactant which was indicated by the reduction in surface tension of the growth medium. Presence of catechol 2, 3-dioxygenase gene in Chry3 indicated its potential for degradation of PAHs through meta cleavage degradation pathway. Both the strains were found to possess catechol 1,2-dioxygenase and catechol 2,3-dioxygenase enzyme activities. Based on morphological and biochemical tests, the cultures were tentatively identified as Bacillus sp. (Chry2) and Pseudomonas sp. (Chry3).

Keywords: Biodegradation - Chrysene - Biosurfactant - Dioxygenase enzyme

Peter O. Abioye[1], A. Abdul Aziz[2] and P. Agamuthu[1]. ([1]Institute of Biological Sciences, University of Malaya, 50603 Kuala Lumpur, Malaysia, [2]Department of Chemical Engineering, University of Malaya, 50603 Kuala Lumpur, Malaysia). **Enhanced Biodegradation of Used Engine Oil in Soil Amended with Organic Wastes. Water, Air, & Soil Pollution, Volume 209(1-4) (2010): 173-179**

Three organic wastes (banana skin (BS), brewery spent grain (BSG), and spent mushroom compost (SMC)) were used for bioremediation of soil spiked with used engine oil to determine the potential of these organic wastes in enhancing biodegradation of used oil in soil. The rates of biodegradation of the oil were studied for a period of 84 days under laboratory conditions. Hydrocarbon-utilizing bacterial counts were high in all the organic waste-amended soil ranging

between 10.2×10^6 and 80.5×10^6 CFU/g compared to unamended control soil throughout the 84 days of study. Oil-contaminated soil amended with BSG showed the highest reduction in total petroleum hydrocarbon with net loss of 26.76% in 84 days compared to other treatments. First-order kinetic model revealed that BSG was the best of the three organic wastes used with biodegradation rate constant of 0.3163 day^{-1} and half-life of 2.19 days. The results obtained demonstrated the potential of organic wastes for oil bioremediation in the order BSG>BS>SMC.

Keywords: Bioremediation - Used engine oil - Organic waste - Hydrocarbon – Bacteria

C. Forbes[a], D. Hughes[a], J. Fox[a], P. Ryan[a] and E. Colleran[a]. ([a] Environmental Microbiology Research Unit, Department of Microbiology, National University of Ireland, Galway, Ireland). High-rate anaerobic degradation of 5 and 6 carbon sugars under thermophilic and mesophilic conditions. Bioresource Technology, Volume 101(11) (2010): 3925-3930

In this research paper, a comparison between thermophilic and mesophilic anaerobic degradation of a variety of the simple sugar components of carbohydrate rich biomass is presented. In order to investigate the degradability of these basic sugars, three synthetic sugar based influents were supplied to two high rate upflow anaerobic hybrid reactors (UAHR) operated at 37 °C (R1) and 55 °C (R2). These influent streams were: d-glucose/sucrose; l-arabinose/d-xylose and l-rhamnose/d-galacturonic acid. The reactors were challenged in terms of influent composition rather than loading rate and were therefore operated at a maximum volumetric loading rate (VLR) of 4.5 gCOD l^{-1} d^{-1} during stable reactor performance. It was found that a switch from a d-glucose/sucrose synthetic influent to an influent composed of l-arabinose/d-xylose resulted in failure of the mesophilic reactor while the thermophilic UAHR was able to tolerate the change of sugar influent at an unchanged VLR of 4.5 gCOD l^{-1} d^{-1}. A subsequent phasing-in approach was used to introduce new sugar influent streams and proved highly successful. The physiology of the biomass was assessed and it was noted that thermophilic anaerobic digestion (AD) involved the formation of acetate and H_2, implying the involvement of homoacetogenic bacteria, while mesophilic AD proceeded via the formation of other intermediates.

Keywords: Thermophilic AD; Pre-hydrolysis; Pectin; Cellulose; OFMSW

Wu-ChungChan[a,] and Yuan-Sheng Lin[a]. ([a] Civil Engineering Department, Chung-Hua University, Hsinchu 30067, Taiwan, ROC). Compounds interaction on the biodegradation of butanol mixture in a biofilter. Bioresource Technology, Volume 101(11) (2010): 4234-4237

Compounds interaction on the biodegradation of *n*-butanol and sec-butanol mixture in a composite bead biofilter was investigated. The biodegradation rate of compounds in the exponential growth phase and stationary phase for the single compound and two compounds mixing systems was determined. The microbial growth rate and biochemical reaction rate of two compounds decreased with increasing compound inlet concentration for the single compound system. The microbial metabolic activity of sec-butanol biodegraded in the microbial growth process and biochemical reaction process was inhibited as *n*-butanol was introduced. This inhibitive effect was more pronounced at higher *n*-butanol inlet concentration and lower sec-butanol inlet concentration for the two compounds mixing system.

Keywords: *n*-Butanol; Sec-butanol; Compound interaction; Biodegradation; Composite bead biofilter

Chen Zhang[b], Le Jia[c], Shenghui Wang[a], Jie Qu[a], Kang Li[a], Lili Xu[a], Yanhua Shi[a] and Yanchun Yan[a]. ([a] Graduate School, Chinese Academy of Agricultural Sciences, Beijing 100081, China, [b] College of Resources and Environmental Sciences, China Agricultural University, Beijing 100094, China, [c] College of Life Sciences, Shandong Agricultural University, Tai'an, Shandong 271018, China). **Biodegradation of beta-cypermethrin by two *Serratia* spp. with different cell surface hydrophobicity. Bioresource Technology, Volume 101(10) (2010): 3423-3429**

Serratia spp. strain JC1 and JCN13, isolated from activated sludge, could degrade and utilize beta-cypermethrin (beta-CP) as the sole carbon and energy sources for growth in the minimal salt media (MSM). The two strains could effectively degrade beta-CP with total inocula biomass 0.1–0.3 g dry wt L^{-1}, at 20–38 °C, pH 6–9, initial beta-CP 25–1000 mg L^{-1} and metabolize it by cleavage of ester and diphenyl ether to yield 3-phenoxybenzoic acid (3-PBA) and phenol, then completely mineralize it. Response surface methodology (RSM) was used to optimize degradation conditions. Under their own optimal degradation conditions, strain JC1 could degrade 92% beta-CP within 10 days and the degradation rate of strain JCN13 reached 89% within 4 days. Cell surface hydrophobicity (CSH) and biodegradation assays indicated that JCN13 has higher hydrophobicity and degradation ability than JC1, and it means the high hydrophobicity of strains could enhance the degradation of beta-CP.

Keywords: Beta-cypermethrin; *Serratia* sp.; Degradation; Cell surface hydrophobicity

Luisa F. González[a], Victor Sarria[b] and Oscar F. Sánchez[a]. ([a] Chemical Engineering Department, Group of Process and Product Design, Universidad de Los Andes, Carrera 1 E No. 19 A 40, Bogotá, Colombia, [b] Chemistry Department, Group of Advanced Oxidation Process for Environmental Remediation, Universidad de Los Andes, Calle 19 A No. 1 – 37 Este, Bogotá, Colombia). **Degradation of chlorophenols by sequential biological-advanced oxidative process using *Trametes pubescens* and TiO_2/UV. Bioresource Technology, Volume 101(10) (2010): 3493-3499**

The degradation of 2-chlorophenol (2-CP), 2,4-dichlorophenol (2,4-DCP), 2,4,6-trichlorophenol (2,4,6-TCP) and pentachlorophenol (PCP) via biological, advanced oxidative process (AOP) and sequential biological-AOP was investigated in this work. The white-rot fungus *Trametes pubescens* was used for the biodegradation of chlorophenols, while in AOP TiO_2/UV was used. In the biological degradation, the effect of glucose as a cofactor was also evaluated. The highest degradations were obtained when the reaction medium was supplemented with glucose, ranging from 94.6% to 37.8%, with degradation activity for 2-CP > 2,4-DCP > PCP > 2,4,6-TCP. During the AOP the removal initial rate increased in the following order 2-CP > 2,4,6-TCP > 2,4-DCP > PCP, and the obtained degradation range from 82.0% to 24.0%. When biological removal process, supplemented with glucose, was followed for an AOP process, 100% degradation was obtained for all the chlorophenols tested. These results suggest that the white-rot fungi *T. pubescens* could be used for the degradation of xenobiotic compounds, and its use with an advanced oxidative process, in a sequential mode, may be considered to obtain a complete removal of them.

Keywords: Chlorophenol; Biodegradation; Photocatalytic degradation; Titanium dioxide; *Trametes pubescens*

Handan Ucun[a], Ergun Yildiz[a] and Alper Nuhoglu[a]. ([a] Department of Environmental Engineering, Engineering Faculty, Ataturk University, Erzurum 25240, Turkey). Phenol biodegradation in a batch jet loop bioreactor (JLB): Kinetics study and pH variation. Bioresource Technology, Volume 101(9) (2010): 2965-2971

Phenol biodegradation in a batch jet loop bioreactor (JLB) using activated sludge was investigated. The biodegradation experiments were conducted at different phenol concentrations (S_0) from 50 to 1000 mg/l. The results of the biodegradation of phenol by JLB show that a good phenol removal of 100%. The biodegradation capacity of the JLB was higher than that of the stirred tank reactor reported in literatures. The Haldane equation was adopted in order to describe the relation between the specific growth rates (μ) and S_0. Kinetic constants of Haldane equation were $\mu_m = 0.119$ 1/h, $K_s = 11.13$ mg/l and $K_i = 250.88$ mg/l. Model equations were simulated using the MATHCAD 7.0 software's ordinary differential equation solver. Simulations were performed at each experiment with different initial phenol concentrations.

Keywords: Phenol; Biodegradation; Jet loop bioreactor (JLB); Haldane equation; Mathematical modeling

G. Mascolo[a], L. Balest[a], D. Cassano[a], G. Laera[a], A. Lopez[a], A. Pollice[a] and C. Salerno[a]. ([a]Istituto di Ricerca Sulle Acque, Consiglio Nazionale delle Ricerche, Viale F. De Blasio, 5, 70123 Bari, Italy). Biodegradability of pharmaceutical industrial wastewater and formation of recalcitrant organic compounds during aerobic biological treatment. Bioresource Technology, Volume 101(8) (2010): 2585-2591

The biodegradability of different wastewater samples originated from the industrial production of three pharmaceuticals (naproxen, acyclovir, and nalidixic acid) was performed through the standard Zahn–Wellens test. Moreover, the wastewater composition before and during the test was evaluated in terms of parent compounds and main metabolites by LC/MS, and the biodegradability of the parent compounds was also assessed by performing extra Zahn–Wellens tests on synthetic solutions. The results, besides showing the relatively good biodegradability of acyclovir and naproxen, evidenced the masking role of the organic matrices, especially in the case of nalidixic acid. The latter compound showed to be recalcitrant and persistent, despite the apparently good performance of the Zahn–Wellens test. Deeper evaluation evidenced that the biodegradation of high concentrations of organic solvents and other biodegradable compound tended to "hide" the lack of removal of the target compound.

Keywords: Biodegradability; Pharmaceuticals; Industrial wastewater; Recalcitrant compounds; Zahn–Wellens test

Awadhesh K. Shukla[a], Pranjali Vishwakarma[a], R.S. Singh[b], S.N. Upadhyay[b] and Suresh K. Dubey[a]. ([a] Department of Botany, Banaras Hindu University, Varanasi 221005, India, [b] Department of Chemical Engineering and Technology, Institute of Technology, Banaras Hindu University, Varanasi 221005, India). Bio-filtration of trichloroethylene using

diazotrophic bacterial community. Bioresource Technology, Volume 101(7) (2010): 2126-2133

Biodegradation of TCE was studied in a biofilter packed with wood charcoal and inoculated with diazotrophic bacterial community isolated from local soil. Steady state TCE removal efficiencies higher than 85% were observed up to inlet load of 2.866 g m^{-3} h^{-1}. The maximum elimination capacity of 5.31 g m^{-3} h^{-1} was observed at an inlet load of more than 7.90 g m^{-3} h^{-1}. The biofilter was sensitive to fluctuations in the process conditions but could easily recover its performance after 10 days shutdown. Almost constant and small pressure drop per unit length and very negligible compaction was observed during the whole experimental period. The molecular analyses such as RT–PCR and gene sequencing revealed the presence of functionally active *Azospirillum* species in the biofilm.

Keywords: TCE degradation; Bio-filtration; Removal efficiency; Diazotroph; *Azospirillum*

Ernest Marco-Urrea[a], Miriam Pérez-Trujillo[b], Paqui Blánquez[a], Teresa Vicent[a] and Gloria Caminal[c]. ([a] Departament d'Enginyeria Química, Institut de Ciència i Tecnologia Ambiental, Universitat Autònoma de Barcelona (UAB), 08193 Bellaterra, Spain, [b] Servei de Ressonància Magnètica Nuclear, UAB, 08193 Bellaterra, Spain, [c] Unitat de Biocatàlisi Aplicada associada al IQAC (CSIC-UAB), Escola d'Enginyeria, UAB, 08193 Bellaterra, Spain). Biodegradation of the analgesic naproxen by *Trametes versicolor* and identification of intermediates using HPLC-DAD-MS and NMR. Bioresource Technology, Volume 101(7) (2010): 2159-2166

The white-rot fungus *Trametes vesicolor* degraded naproxen (10 mg L^{-1}) in a liquid medium to non-detectable levels after 6 h. When naproxen was added in the range of concentrations typically found in the environment (55 µg L^{-1}), it was almost completely degraded (95%) after 5 h. *In vitro* degradation experiments with purified laccase and purified laccase plus mediator 1-hydroxybenzotriazol showed slight and almost complete naproxen degradation, respectively. A noticeable inhibition on naproxen degradation was also observed when the cytochrome P450 inhibitor 1-aminobenzotriazole was added to the fungal cultures. These data suggest that both enzymatic systems could play a role in naproxen degradation. 2-(6-hydroxynaphthalen-2-yl) propanoic acid and 1-(6-methoxynaphthalen-2-yl)ethanone were structurally elucidated by HPLC-DAD-MS and NMR as degradation intermediates of naproxen. After 6 h of incubation, both parent compound and intermediates disappeared from the medium. The non-toxicity of the treated medium was confirmed by Microtox test.

Keywords: Naproxen; *Trametes versicolor*; NMR; Cytochrome P450; Laccase

A.J. Ward[a] and M.S. Kumar[a]. ([a] South Australian Research and Development Institute (SARDI), Integrated Bio-Systems Roseworthy, Australia). Bio-conversion rate and optimum harvest intervals for *Moina australiensis* using digested piggery effluent and *Chlorella vulgaris* as a food source. Bioresource Technology, Volume 101(7) (2010) :2210-2216

The bio-conversion rate of *Moina australiensis* fed with *Chlorella vulgaris* grown on digested piggery effluent at three different feeding rates was determined and a 2, 3 and 4-day harvest interval strategy was investigated. This study indicates that *C. vulgaris* is a suitable food source for *M. australiensis*. A significant difference ($P \leqslant 0.001$) in the feeding rate against mean total

populations was found among treatments. The increase in the amount of algae fed accelerated the production rate, and the population density peaked faster in the high *C. vulgaris* fed treatment. The BCR calculated from this experiment indicates that for every 1000 mg of *C. vulgaris* fed there was an increase of 437.9 mg of *M. australiensis* biomass produced. A significant difference ($P \leqslant 0.001$) in biomass production among the different harvest interval treatments was observed. The 2-day harvest interval treatment produced 7.78 g of *M. australiensis* followed by 6.89 g in the 3 day and 5.01 g in the 4-day harvest interval treatment. This study provides strong evidence that *M. australiensis* can utilise the bacterial blooms and bio-films associated with digested piggery effluent as a food source.

Keywords: *Moina australiensis*; *Chlorella vulgaris*; Piggery effluent; Bio-conversion rate; Harvest interval

A.R. Khataee[a], G. Dehghan[b], A. Ebadi[a, b], M. Zarei[a] and M. Pourhassan[b]. ([a]Department of Applied Chemistry, Faculty of Chemistry, University of Tabriz, Tabriz, Iran, [b] Department of Biology, Faculty of Science, University of Tabriz, Tabriz, Iran). **Biological treatment of a dye solution by Macroalgae *Chara* sp.: Effect of operational parameters, intermediates identification and artificial neural network modeling. Bioresource Technology, Volume 101(7) (2010): 2252-2258**

The potential of a macroalgae *Chara* sp. was investigated as a viable biomaterial for biological treatment of Malachite Green (MG) solution. The effects of operational parameters such as temperature, pH, initial dye concentration, reaction time and amount of algae on biological decolorization efficiency were studied. Biological treatment of MG solution by live and dead algae was compared. The reusability and efficiency of the live algae in long-term repetitive operations were also examined. The batch experiments results revealed the ability of algal species in biological degradation of the dye. The biological degradation compounds formed in this process were analyzed by UV–Vis, FT-IR and GC-Mass techniques. The degradation pathway of MG was proposed based on the identified compounds. In addition, an artificial neural network model was developed to predict the biological degradation efficiency. The findings indicated that ANN provides reasonable predictive performance ($R^2 = 0.970$). The influence of each parameter on the variable studied was assessed, reaction time being the most significant factor, followed by temperature of the solution.

Keywords: Biodegradation; Macroalgae; Malachite Green; Modeling; Decolorization

Carlos E. Rodríguez-Rodríguez[a], Ernest Marco-Urrea[b] and Gloria Caminal[a]. ([a]Unitat de Biocatàlisi Aplicada associada al IQAC (CSIC-UAB), Escola d'Enginyeria, UAB, 08193 Bellaterra, Spain, [b] Departament d'Enginyeria Química and Institut de Ciència i Tecnologia Ambiental, Universitat Autònoma de Barcelona (UAB), 08193 Bellaterra, Spain). **Degradation of naproxen and carbamazepine in spiked sludge by slurry and solid-phase *Trametes versicolor* systems. Bioresource Technology, Volume 101(7) (2010): 2259-2266**

Growth and activity of the white-rot fungus *Trametes versicolor* on sewage sludge were assessed in bioslurry and solid-phase systems. Bioslurry cultures with different loads of sludge (10%, 25% and 38%, w/v) were performed. A lag phase of at least 2 d appeared in the 25 and 38%-

cultures, however, the total fungal biomass was higher for the latter and lower for the 10%-culture after 30 d, as revealed by ergosterol determination. Detectable laccase activity levels were found in the 10 and 25%-cultures (up to 1308 and 2588 AU L^{-1}, respectively) while it was negligible in the 38%-culture. Important levels of ergosterol and laccase were obtained over a 60 d period in sludge solid-phase cultures amended with different concentrations of wheat straw pellets as lignocellulosic bulking material. Degradation experiments in 25%-bioslurry cultures spiked with naproxene (NAP, analgesic) and carbamazepine (CBZ, antiepileptic) showed depletion of around 47% and 57% within 24 h, respectively. Complete depletion of NAP and around 48% for CBZ were achieved within 72 h in sludge solid cultures with 38% bulking material. CBZ degradation is especially remarkable due to its high persistence in wastewater treatment plants. Results showed that *T. versicolor* may be an interesting bioremediation agent for elimination of emerging pollutants in sewage sludge.

Keywords: *Trametes versicolor*; Sewage sludge; Bioslurry; Solid-phase fermentation; Emerging pollutants

Xia Jiang[a] and Joo Hwa Tay[a]. ([a] School of Civil and Environmental Engineering, Nanyang Technological University, Blk N1, 50 Nanyang Avenue, Singapore 639798, Singapore). **Microbial community structures in a horizontal biotrickling filter degrading H_2S and NH_3. Bioresource Technology, Volume 101(6) (2010): 1635-1641**

In this study, the temporal and spatial differences in the microbial community structures in a horizontal biotrickling filter (HBTF) for simultaneous biodegradation of H_2S and NH_3 was investigated by denaturing gradient gel electrophoresis (DGGE) of polymerase chain reaction (PCR) amplified 16S rRNA gene fragments. Over 90% H_2S and 95% NH_3 were removed simultaneously under the loadings up to 137 g m^{-3} h^{-1} in most cases. PCR–DGGE fingerprints revealed high population diversity of the biofilm in the HBTF. The microbial community structures in the HBTF could adapt to changing operating conditions, e.g., inlet loadings and substrates starvation, while they were relatively stable under the constant loadings. In addition, the microbial community structures differed as a function of the section of the HBTF during early operational period, while they were much more similar over extended operation.

Keywords: Microbial community structures; Denaturing gradient gel electrophoresis; Biotrickling filter; Hydrogen sulfide; Ammonia

Luis A. Sayavedra-Soto[1], Barbara Gvakharia[1], Peter J. Bottomley[2], Daniel J. Arp[1] and Mark E. Dolan[3]. ([1]Department of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97331, USA, [2]Department of Microbiology, Oregon State University, Corvallis, OR 97331, USA, [3]School of Chemical, Biological and Environmental Engineering, Oregon State University, Corvallis, OR 97331, USA). **Nitrification and degradation of halogenated hydrocarbons—a tenuous balance for ammonia-oxidizing bacteria. Applied Microbiology and Biotechnology, Volume 86(2) (2010): 435-444**

The process of nitrification has the potential for the in situ bioremediation of halogenated compounds provided a number of challenges can be overcome. In nitrification, the microbial process where ammonia is oxidized to nitrate, ammonia-oxidizing bacteria (AOB) are key players and are capable of carrying out the biodegradation of recalcitrant halogenated compounds. Through industrial uses, halogenated compounds often find their way into wastewater, contaminating the environment and bodies of water that supply drinking water. In

the reclamation of wastewater, halogenated compounds can be degraded by AOB but can also be detrimental to the process of nitrification. This minireview considers the ability of AOB to carry out cometabolism of halogenated compounds and the consequent inhibition of nitrification. Possible cometabolism monitoring methods that were derived from current information about AOB genomes are also discussed. AOB expression microarrays have detected mRNA of genes that are expressed at higher levels during stress and are deemed "sentinel" genes. Promoters of selected "sentinel" genes have been cloned and used to drive the expression of gene-reporter constructs. The latter are being tested as early warning biosensors of cometabolism-induced damage in *Nitrosomonas europaea* with promising results. These and other biosensors may help to preserve the tenuous balance that exists when nitrification occurs in waste streams containing alternative AOB substrates such as halogenated hydrocarbons.

Keywords: Nitrification - Chlorinated aliphatic hydrocarbons - *N. europaea* - Degradation – Cometabolism

Nai-Dong Chen[1], Jian Zhang[1], Ji-Hua Liu[2] and Bo-Yang Yu[2]. ([1]Department of Complex Prescription of TCM, China Pharmaceutical University, 639 Long Mian Road, Nanjing, 211198, People's Republic of China, [2]Key Laboratory of Modern Chinese Medicines, Ministry of Education, China Pharmaceutical University, 24 Tong Jia Xiang, Nanjing, 210038, People's Republic of China). **Microbial conversion of ruscogenin by *Gliocladium deliquescens* NRRL1086: glycosylation at C-1. Applied Microbiology and Biotechnology, Volume 86(2) (2010): 491-497**

The glycosylation of ruscogenin (**1**) by *Gliocladium deliquescens* NRRL 1086 was observed and gave a regioselectively glycosylated product identified as ruscogenin 1-O-β-D-glucopyranoside (**2**) by infrared, mass spectrometry, and nuclear magnetic resonance spectra. Time-course studies indicated that it appeared to be favorable to accumulate **2** when ruscogenin was added to the 24-h-old stage II culture, and the yield of **2** was about 20.1% during 120168 h. It was noted that additional carbohydrates could significantly increase glycoside formation and the yield of **2** even reached as high as 68% compared with the control 20.1%. The primary investigation about the characteristics of the enzyme resulted that the reaction was blocked by β-glycosidase inhibitor imidazole, however, was enhanced remarkably by glycosyltransferase inhibitor sodium dodecyl sulfate. To our knowledge, this is the first reported case of producing steroidal saponin by microbial transformation, and *G. deliquescens* NRRL1086 would be a practical and highly efficient tool in producing natural ruscogenin monoside.

Keywords: *Gliocladium deliquescens* NRRL1086 - Biotransformation - Ruscogenin - Microbial glycosylation

Jasperien De Weert[1, 2], Marc Viñas[3], Tim Grotenhuis[2], Huub Rijnaarts[1, 2] and Alette Langenhoff[1]. ([1]Deltares, Princetonlaan 6, 3584 CB Utrecht, The Netherlands, [2]Section of Environmental Technology, Wageningen University, Bomenweg 2, 6703 HD Wageningen, The Netherlands, [3]GIRO Technological Center, Rambla Pompeu Fabra 1. E-08100 Mollet del Vallès, Barcelona, Spain). **Aerobic nonylphenol degradation and nitro-nonylphenol formation by microbial cultures from sediments. Applied Microbiology and Biotechnology, Volume 86(2) (2010): 761-771**

Nonylphenol (NP) is an estrogenic pollutant which is widely present in the aquatic environment. Biodegradation of NP can reduce the toxicological risk. In this study, aerobic biodegradation of NP in river sediment was investigated. The sediment used for the microcosm experiments was aged polluted with NP. The biodegradation of NP in the sediment occurred within 8 days with a lag phase of 2 days at 30°C. During the biodegradation, nitro-nonylphenol metabolites were formed, which were further degraded to unknown compounds. The attached nitro-group originated from the ammonium in the medium. Five subsequent transfers were performed from original sediment and yielded a final stable population. In this NP-degrading culture, the microorganisms possibly involved in the biotransformation of NP to nitro-nonylphenol were related to ammonium-oxidizing bacteria. Besides the degradation of NP via nitro-nonylphenol, bacteria related to phenol-degrading species, which degrade phenol via ring cleavage, are abundantly present.

Keywords: Nonylphenol - Nitro-nonylphenol - Aerobic degradation - *Nitrosomonas*

Petra Bombach[1, 2], Hans H. Richnow[1], Matthias Kästner[3] and Anko Fischer[1, 2]. ([1]Department of Isotope Biogeochemistry, UFZ - Helmholtz Centre for Environmental Research, Permoserstrasse 15, 04318 Leipzig, Germany, [2]Isodetect - Company for Isotope Monitoring (Branch Leipzig), Permoserstrasse 15, 04318 Leipzig, Germany, [3]Department of Environmental Biotechnology, UFZ - Helmholtz Centre for Environmental Research, Permoserstrasse 15, 04318 Leipzig, Germany). **Current approaches for the assessment of in situ biodegradation. Applied Microbiology and Biotechnology, Volume 86(3) (2010): 839-852**

Considering the high costs and technical difficulties associated with conventional remediation strategies, in situ biodegradation has become a promising approach for cleaning up contaminated aquifers. To verify if in situ biodegradation of organic contaminants is taking place at a contaminated site and to determine if these processes are efficient enough to replace conventional cleanup technologies, a comprehensive characterization of site-specific biodegradation processes is essential. In recent years, several strategies including geochemical analyses, microbial and molecular methods, tracer tests, metabolite analysis, compound-specific isotope analysis, and in situ microcosms have been developed to investigate the relevance of biodegradation processes for cleaning up contaminated aquifers. In this review, we outline current approaches for the assessment of in situ biodegradation and discuss their potential and limitations. We also discuss the benefits of research strategies combining complementary methods to gain a more comprehensive understanding of the complex hydrogeological and microbial interactions governing contaminant biodegradation in the field.

Keywords: Microbial in situ degradation - Contaminated aquifer - In situ microcosm - Compound-specific isotope analysis - Metabolites - Functional genes

Yongming Zhang[1], Lei Wang[1] and Bruce E. Rittmann[2]. ([1]Department of Environmental Engineering, College of Life and Environmental Science, Shanghai Normal University, Shanghai, 200234, People's Republic of China, [2]Center for Environmental Biotechnology, Biodesign Institute, Arizona State University, Tempe, AZ 85287-5801, USA). **Integrated photocatalytic-biological reactor for accelerated phenol mineralization. Applied Microbiology and Biotechnology, Volume 86(6) (2010): 1977-1985**

An integrated photocatalytic-biological reactor (IPBR) was developed for accelerated phenol degradation and mineralization. In the IPBR, photodegradation and biodegradation occurred simultaneously, but in two separated zones: a piece of mat-glass plate coated with TiO_2 film and illuminated by UV light was connected by internal circulation to a honeycomb ceramic that was the biofilm carrier for biodegradation. This arrangement was designed to give intimate coupling of photocatalysis and biodegradation. Phenol degradation was investigated by following three protocols: photocatlysis with TiO_2 film under ultraviolet light, but no biofilm (photodegradation); biofilm biodegradation with no UV light (biodegradation); and simultaneous photodegradation and biodegradation (intimately coupled photobiodegradation). Photodegradation alone could partly degrade phenol, but was not able to achieve significant mineralization, even with an HRT of 10 h. Biodegradation alone could completely degrade phenol, but it did not mineralize the COD by more than 74%. Photobiodegradation allowed continuous rapid degradation of phenol, but it also led to more complete mineralization of phenol (up to 92%) than the other protocols. The results demonstrate that intimate coupling was achieved by protecting the biofilm from UV and free-radical inhibition. With phenol as the target compound, the main advantage of intimate coupling in the IPBR was increased mineralization, presumably because photocatalysis made soluble microbial products more rapidly biodegradable.

Keywords: Biofilm - Photocatalysis - Bioreactor - Wastewater treatment - Phenol

Hosseini Farzaneh[1]*, Malekzadeh Fereidon[1], Amirmozafari Noor[2] and Ghaemi Naser[3]. ([1]Department of Biology, College of Basic Science, Islamic Azad University Research and Science Tehran branch, Tehran, Iran, [2]Department of microbiology, Iran University of Medical Sciences, Tehran, Iran, [3]Department of Biotechnology, College of Science, Tehran University, Tehran, Iran. *Corresponding author. E-mail: farzaneh953@yahoo.com. Tel: +98021-22641842). **Biodegradation of dodecylbenzene solfonate sodium by *Stenotrophomonas maltophilia* Biofilm. African Journal of Biotechnology Vol. 9 (1) (2010): 055-062**

Immobilization for microbial cultures has proved to be advantageous in municipal and industrial sewage treatment because of high degradation efficiency and good operational stability. In this survey, a bacterial strain was isolated from activated sludge that utilized branched anionic surfactants (BAS) as a sole carbon source. Identification of isolated strain was from 16S rRNA sequencing method. The immobilized cells on silanized glass beads as support and unmodified surfaces were used for removal of BAS; both types showed effective biodegrading of BAS. The removal rate in silanized surface was about 2 fold of unmodified surface. The result of biodegradation was studied by HPLC method and scanning electron microscope.

Keywords: Branched anionic surfactants, biofilm, biodegradation, silanization.

Mathabatha Evodia Setati. (Discipline of Microbiology, School of Biochemistry, Genetics and Microbiology, University of KwaZulu-Natal, P/Bag X54001, Durban 4000, South Africa. E-mail: setatim@ukzn.ac.za. Fax: +27(31) 260 7809. Tel: +27(31) 260 7405). **Diversity and industrial potential of hydrolase-producing halophilic/halotolerant eubacteria. African Journal of Biotechnology Vol. 9 (11) (2010): 1555-1560**

Halophilic and haloterant eubacteria have been isolated from different marine and hypersaline environments. Halophilic eubacteria also occur in environments typified by more than one soda lakes which are both hypersaline and extremely alkaline. These organisms have been shown to produce a wide array of hydrolytic enzymes including proteases, amylases, xylanases, cellulases as well as lipases and DNases. These enzymes are commonly applied in the production of fermented food and food supplements, in animal feed, laundry detergents and textile industries. Several studies have shown that enzymes derived from halophilic and halotolerant eubacteria are not only halostable but may also be thermostable and alkalistable. This extremophilicity make the enzymes suitable candidates in various fields of biotechnology and may even open up new application opportunities.

Keywords: Halophilic/halotolerant, Hydrolases, biodiversity, alkaliphilic, proteases, amylases, xylanases, cellulases.

Xue-Qin Tao[1], Jie-Ping Liu[1], Gui-Ning Lu[2]*, Xiu Guo[1], Hui-Ping Jiang[1] and Guan-Qiu Sun[1]. ([1]School of Environmental Science and Engineering, Zhongkai University of Agriculture and Engineering, Guangzhou 510225, P. R. China, [2]School of Environmental Science and Engineering, South China University of Technology, Guangzhou Higher Education Mega Center, Guangzhou 510006, P. R. China. *Corresponding author. E-mail: lutao@scut.edu.cn. Tel/ Fax: +86-20-89003188.). **Biodegradation of phenanthrene in artificial seawater by using free and immobilized strain of *Sphingomonas* sp. GY2B. African Journal of Biotechnology Vol. 9 (18) (2010): 2654-2660**

Biodegradation has been suggested as an alternative way to remove polycyclic aromatic hydrocarbons (PAHs) from contaminated environment. Phenanthrene is a representative carcinogenic PAHs containing “bay-region” and “K-region”. Strain *Sphingomonas* sp. GY2B is a high efficient phenanthrene-degrading strain isolated from crude oil contaminated soils and had a broad-spectrum degradation ability on PAHs and related aromatic compounds. This paper reports the domestication of strain *Sphingomonas* sp. GY2B in artificial seawater (AS) and the immobilization of the strain onto rice straw. Results showed that adding 85% artificial seawater had very low impact on the growth and phenanthrene degradation ability of strain GY2B being domesticated for five generations. Phenanthrene was rapidly degraded when the growth of strain GY2B was in the exponential phase that the initial added 100 mgL^{-1} phenanthrene had been almost completely degraded within 66 h. The optimal immobilization carrier weight and length of rice straw were 25 gL^{-1} and 0.5 cm, respectively. The immobilized strain GY2B had high degradation rate both in mineral salts medium and 80% artificial seawater, and was higher than that of the free strain GY2B. More than 95% phenanthrene (100 mgL^{-1}) was degraded within 32 h, and the phenanthrene degradation percentages were > 99.5% after 67 h for immobilized strains. Immobilization of strain GY2B with rice straw possesses a good application potential in the treatment of wastewater and bioremediation of estuary and offshore environment contaminated by phenanthrene.

Keywords: Artificial seawater, biodegradation, immobilization, phenanthrene, polycyclic aromatic hydrocarbons, rice straw, *Sphingomonas* sp. GY2B.

Abbreviations: **PAHs**, polycyclic aromatic hydrocarbons; **MSM**, mineral salts medium; **AS**, artificial seawater; **PDR**, phenanthrene degradation rate.

K. Sarayu[1] and S. Sandhya[1]. ([1]National Environmental Engineering Research Institute, CSIR Complex, Chennai, 600113, India). Aerobic Biodegradation Pathway for Remazol Orange by *Pseudomonas aeruginosa.* Applied Biochemistry and Biotechnology, Volume 160(4) (2010): 1241-1253

Removal of azo dyes from effluent generated by textile industries is rather difficult. Azo dyes represent a major class of synthetic colorants that are mutagenic and carcinogenic. *Pseudomonas aeruginosa* grew well in the presence of Remazol Orange (RO) and was able to decolorize and degrade it. In the present study, the decolorization and degradation efficiency using single culture *P. aeruginosa* with RO and textile wastewaters is studied. The elucidation of decolorization pathway for *P. aeruginosa* is of special interest. The degradation pathway and the metabolic products formed during the degradation were also predicted with the help of high performance liquid chromatography, Fourier transform infrared spectroscopy, and nuclear magnetic resonance spectroscopy analysis. The data show the cleavage of the azo dye RO to form both methyl metanilic acid and 4-aminobenzoic acid after decolorization and finally to oxidation forms benzoic acid, alkenes, aldehydes, and alkynes. The organism was able to decolorize the dye RO and wastewater effectively to the maximum of 82.4% and 62%, respectively.

Keywords: *Pseudomonas aeruginosa* - Decolorization - Biodegradation - Metabolic pathway - Textile wastewater treatment - Growth kinetics

Daljit Singh Arora[1] and Rakesh Kumar Sharma[1]. ([1]Department of Microbiology, Microbial Technology Laboratory, Guru Nanak Dev University, Amritsar, 143005, Punjab, India). Ligninolytic Fungal Laccases and Their Biotechnological Applications. Applied Biochemistry and Biotechnology, Volume 160(6) (2010): 1760-1788

Lignin is amorphous in nature, lacks stereoregularity, and is not susceptible to hydrolytic attack. Despite its resistant nature, it is however degraded by various microorganisms, particularly, white-rot fungi. Such fungi are capable of extracellular production of lignin peroxidase, manganese peroxidase, and laccase, the three major enzymes associated with ligninolysis. Though all white-rot fungi do not produce all the three enzymes, laccase occupies an important place in ligninolysis. Laccase belongs to a diverse group of enzymes called oxidoreductases and is also known as benzenediol: oxygen oxidoreductase. They have low substrate specificity. The copper-containing enzyme laccase has been detected in a variety of organisms such as bacteria, fungi, plants, and insects. Mostly, these are extracellular proteins, although intracellular laccases have also been detected in some fungi and insects. Fungal laccases are believed to play a variety of roles, such as, morphogenesis, pathogenesis, and lignin degradation. As an oxidase, laccase is used in many agricultural, industrial, and medicinal applications. Current investigations are focused on laccase-based biooxidation, biotransformation, biosensor, and enzymatic synthesis of organic compounds. By enhancing laccase production using different physiochemical parameters, better understanding of the mechanism for the reactions of interest, and optimizing the catalytic activity of laccase, it can be used in a better way in diverse fields of biotechnology.

Keywords: Bioremediation - Laccase - Lignin - Lignocellulosics - White-rot fungi

Zhengbo Yue, Charles Teater, Yan Liu, James MacLellan, Wei Liao. * (Department of Biosystems and Agricultural Engineering, Michigan State University, 202 Farrall Hall, East Lansing, Michigan 48824; telephone: 517-432-7205; fax: 517-432-2892). **A sustainable pathway of cellulosic ethanol production integrating anaerobic digestion with biorefining. Biotechnology and Bioengineering, Volume 105(6) (2010): 1031 - 1039**

Anaerobic digestion (AD) of animal manure is traditionally classified as a treatment to reduce the environmental impacts of odor, pathogens, and excess nutrients associated with animal manure. This report shows that AD also changes the composition of manure fiber and makes it suitable as a cellulosic feedstock for ethanol production. Anaerobically digested manure fiber (AD fiber) contains less hemicellulose (11%) and more cellulose (32%) than raw manure, and has better enzymatic digestibility than switchgrass. Using the optimal dilute alkaline pretreatment (2% sodium hydroxide, 130°C, and 2 h), enzymatic hydrolysis of 10% (dry basis) pretreated AD fiber produces 51 g/L glucose at a conversion rate of 90%. The ethanol fermentation on the hydrolysate has a 72% ethanol yield. The results indicate that 120 million dry tons of cattle manure available annually in the U.S. can generate 63 million dry tons of AD fiber that can produce more than 1.67 billion gallons of ethanol. Integrating AD with biorefining will make significant contribution to the cellulosic ethanol production.

Keywords: anaerobic digestion • cattle manure • ethanol • lignocellulose

Pedro A. Isaza, Andrew J. Daugulis .* (Department of Chemical Engineering, Queen's University, Kingston, Ontario, Canada K7L 3N6; telephone: 613-533-2784; fax: 613-533-6637). **Enhanced degradation of phenanthrene in a solid-liquid two-phase partitioning bioreactor via sonication. Biotechnology and Bioengineering, Volume 105 (5) (2010): 997 - 1001**

The current article examined the feasibility of inducing improved delivery and degradation of phenanthrene in a solid-liquid partitioning bioreactor system at bench scale by means of ultrasonic energy input. Initial degradation rates of phenanthrene by a microbial consortium, delivered from Desmopan, were improved 2.7-fold in the presence of sonication relative to unsonicated controls. Results demonstrated that an operating window involving on/off sonication cycling improved substrate delivery and rational selection of ultrasound cycling profiles could lead to even further enhancements. Additionally, all results were obtained in a conventional bioreactor with commercial ultrasonic equipment and a commercially available polymer. Subsequent DGGE analysis demonstrated that the sonication cycles selected maintained consortium compositions, relative to control cases, and suggest that exposure would not reduce degradative capabilities under the periods of irradiation examined. Finally, consortium members were identified as belonging to the *Pandoraea*, *Sphingobium*, and *Pseudoxanthomonas* genera. Comparison of genetic sequences in the Ribosomal Database Project revealed that some of the bacterial members, identified at the strain level, had been previously observed in PAH degradations, while others have been reported only in the degradation of other aromatics, such as pesticides.

Keywords: sonication • solid-liquid two-phase partitioning bioreactors • biodegradation • phenanthrene • enhanced delivery

Biosensor

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Most national standards for assessment of water quality include physical and chemical indicators relevant to specific pollutants and stressors. However, biological communities reflect not only current conditions of aquatic resources but also change in conditions over time and impacts from multiple stressors. Assessing the health of the aquatic community (that is, bioassessments) has proven to be critical in protecting and maintaining healthy surface waters under the mandates of regulatory frameworks, such as the Clean Water Act in the USA and the Water Framework Directive of the European Union. Whereas, in Thailand water standards, bioassessment is lacking in favor of chemical criteria, only coliform bacteria measurement can be considered a surrogate biological parameter. Our paper argues that incorporating bioassessment will improve water resource condition evaluations and recommends the use of the benthic macroinvertebrate assemblage as a bioassessment framework in Thailand. We discuss the implementation of a bioassessment program that consists of two major components, (a) a scientifically valid technical approach and (b) consideration of technical resources for a cost-effective program. The technical design comprises (1) classification of streams into similar groupings, (2) design of a biological survey, (3) a well-documented sampling protocol, (4) calibration of biological metrics for data analysis, (5) development of criteria for determination of ecological condition, and (6) communication of the results to citizens and policymakers. A cost-effective way to develop a bioassessment program that will improve Thailand's ability to measure water quality and to make good decisions to attain healthy quality status is to establish partnerships by coordinating efforts and sharing data and technology with adjacent regional environmental offices or provinces. This collaboration would be fostered through a long-term national water resources management strategy and clear definition of goals and desired outcomes that are critical components of the overall National Plan for Ecological Health.

Keywords: Water quality - Biological indicators - Benthic macroinvertebrates - Thai streams - Water resource management - Bioassessment

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The distribution and potential bioaccumulation of dietary and waterborne cadmium and lead in tissues of sea bream (*Sparus aurata*), a major aquaculture species, was studied in relation to three different fish farming systems. Metallothionein levels in fish tissues were also evaluated. Results demonstrate that metal concentrations in various tissues significantly vary among fish culture systems. Different tissues show different capacity for accumulating heavy metals. The content of both cadmium and lead is not strictly correlated with that of metallothionein. Indeed, the marked accumulation of both metals in liver, as well as the high lead content found in gills and kidney, are not accompanied by a concomitant accumulation of metallothioneins in these tissues. No correlation is present between heavy metals and metallothionein content in muscle tissue. The results also demonstrate that cadmium accumulates mainly via dietary food, whereas lead accumulation is not of food origin. Noteworthy is that the concentration of the two metals found in muscle in all instances is lower than the limits established by European Union legislation for fish destined for human consumption.

Keywords: Cadmium - Lead - Tissue distribution - Metallothionein - Aquaculture - Sea bream

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Indicators were devised to classify air pollution monitoring sites according to the type of expected photochemical pollution. The indicators are based on measured ozone volume fractions, the most frequently monitored component of photochemical pollution, and in particular on two contributions: one due to the ratio of daily maximum-to-minimum ozone volume fractions and the other to observed peak values. The two contributions regarded as independent are logically connected by "and" and therefore mathematically combined by multiplication. The criterion of classification is mainly described by the mentioned ratio and incidences of ozone volume fractions exceeding the limit of 80 ppb. Twelve monitoring stations within the European network (Cooperative programme for monitoring and evaluation of long-range transmission of air pollutants in Europe, EMEP) were classified according to this indicator predicting what ozone levels can be expected at the particular sites during the growth season (April through September) into three groups: clean, medium, and polluted, based on the data for the 7 years (1997 to 2003).

Keywords: Tropospheric ozone - Photochemical pollution - Air pollution indicator - Monitoring - EMEP

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Normal University, Beijing, 100875, People's Republic of China, [2]Institute of Agricultural Resources and Regional Planning, Chinese Academy of Agricultural Sciences, Beijing, 100081, People's Republic of China). Functional bacteria as potential indicators of water quality in Three Gorges Reservoir, China. Environmental Monitoring and Assessment, Volume 163(1-4) (2010): 607-617

The distributions of culturable bacteria and functional bacteria associated with nitrogen (N) or phosphorus (P) in the backwater areas of the Three Gorges Reservoir (TGR) were investigated. Results from seven locations in the TGR indicated that the abundance of total bacteria was high, with 8.12 Ã— 10^6, 2.70 Ã— 10^7, and 6.73 Ã— 10^{10} colony-forming units per milliliter or per gram dry weight in surface water, bottom water, and sediments, respectively. Aquatic environments with higher nutrient loadings possessed higher bacteria densities and lower bacteria community diversities. Eight kinds of functional bacteria ratios, including surface water to bottom water and ratios of water to sediments, were calculated, in which four kinds of functional bacteria, namely, nitrogen-fixing bacteria, ammonia-oxidizing bacteria, nitrite-oxidizing bacteria, and organophosphate-solubilizing bacteria, displayed obvious differences in different locations. Based on the functional bacteria ratios of water to sediments, it was found that the obtained result of location grouping from cluster analysis was similar to that based on the community-level physiological profiles studies. The above results showed that the ratios of functional bacteria could distinguish the aquatic environments with different trophic conditions in the TGR. This demonstrated that the distribution ratios of functional bacteria in aquatic environments could work as potential bioindicators to reflect the trophic condition of the water.

Keywords: Bacteria abundance - Eutrophication - Functional bacteria - Microbial community diversity - Three Gorges Reservoir

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Diffuse Nitrogen (N) loss from agriculture is a major factor contributing to increased concentrations of nitrate in surface and groundwater, and of N_2O and NH_3 in the atmosphere. Different approaches to assess diffuse N losses from agriculture have been proposed, among other direct measurements of N loads in leachate and groundwater, and physically-based modelling. However, both these approaches have serious drawbacks and are awkward to use at a routine base. N loss indicators (NLIs) are environmental management tools for assessing the risk of diffuse N losses from agricultural fields. They range in complexity from simple proxy variables to elaborate systems of algebraic equations. Here we present an overview of NLIs developed in different parts of the world. NLIs can be categorized into source-based, transport-based, and composite approaches. Several issues demand more attention in future studies. (1) Is incorporation of leaching losses and gaseous losses into one single NLI warranted? (2) Is it sufficient to restrict the focus on the rooted soil zone without considering the vadose zone and aquifer? (3) Calibration and validation of NLIs using field data of N loss seems not sufficient. Comparisons of several different NLIs with each other needs more attention; however, the different scaling of NLIs impedes comparability. (4) Sensitivity of input parameters with regard

to the final NLI output needs more attention in future studies. (5) For environmental management purposes, factors addressing management decision by farmers deserve more attention.

Keywords: Environmental indicator - Agricultural nutrient management - Risk assessment - Nitrate leaching - Nitrogen loss indicator - Non-point source pollution

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Pesticide Risk Indicators (PRIs) are widely used to evaluate and compare the potential health and environmental risks of pesticide use and to guide pest control policies and practices. They are applied to agricultural, landscape and structural pest management by governmental agencies, private institutions and individuals. PRIs typically assess only the potential risks associated with the active ingredients because, with few exceptions, pesticide manufacturers disclose only the identity of the active ingredients which generally comprise only a minor portion of pesticide products. We show that when inert ingredients are identified and assessed by the same process as the active ingredient, the product specific risk can be much greater than that calculated for the active ingredient alone. To maintain transparency in risk assessment, all those who develop and apply PRIs or make decisions based on their output, should clearly disclose and discuss the limitations of the method.

Keywords: Pesticide Risk Indicators - Inert ingredients - EIQ - GUS

Aloysio da S. Ferrão-Filho[a], Maria Carolina S. Soares[b], Valéria Freitas de Magalhães[c], and Sandra M.F.O. Azevedo[c]. ([a] Laboratório de Avaliação e Promoção da Saúde Ambiental, Departamento de Biologia, Instituto Oswaldo Cruz, FIOCRUZ, Av. Brasil 4365, Manguinhos, Rio de Janeiro, RJ 21045-900, Brazil, [b] Departamento de Engenharia Sanitária e Ambiental Faculdade de Engenharia, Universidade Federal de Juiz de Fora, Juiz de Fora, MG 36036-900, Brazil, [c] Laboratório de Ecofisiologia e Toxicologia de Cianobactérias, Instituto de Biofísica Carlos Chagas Filho, CCS, Universidade Federal do Rio de Janeiro, Ilha do Fundão, Rio de Janeiro, RJ 21949-900, Brazil). **A rapid bioassay for detecting saxitoxins using a *Daphnia* acute toxicity test. Environmental Pollution, Volume 158(6) (2010): 2084-2093**

Bioassays using *Daphnia pulex* and *Moina micrura* were designed to detect cyanobacterial neurotoxins in raw water samples. Phytoplankton and cyanotoxins from seston were analyzed during 15 months in a eutrophic reservoir. Effective time to immobilize 50% of the exposed individuals (ET_{50}) was adopted as the endpoint. Paralysis of swimming movements was observed between ~0.5–3 h of exposure to lake water containing toxic cyanobacteria, followed by an almost complete recovery of the swimming activity within 24 h after being placed in control water. The same effects were observed in bioassays with a saxitoxin-producer strain of *Cylindrospermopsis raciborskii* isolated from the reservoir. Regression analysis showed significant relationships between ET_{50} *vs.* cell density, biomass and saxitoxins content, suggesting that the paralysis of *Daphnia* in lake water samples was caused by saxitoxins found

in *C. raciborskii. Daphnia* bioassay was found to be a sensitive method for detecting fast-acting neurotoxins in natural samples, with important advantages over mouse bioassays.

A new *Daphnia* bioassay, as an alternative to the mouse bioassay, is able to detect effects of fast-acting, potent neurotoxins in raw water.

Keywords: Cyanobacteria; Cladocerans; *Daphnia*; Saxitoxins; *Cylindrospermopsis*

Xavier Laffray[a], Christophe Rose[a] and Jean-Pierre Garrec[a]. ([a] UMR INRA/UHP Forest Ecology and Ecophysiology, Phytoecology team, Atmospheric Pollution Laboratory, INRA Nancy, Route de l'Arboretum, F-54280 Champenoux, France). **Biomonitoring of traffic-related nitrogen oxides in the Maurienne valley (Savoie, France), using purple moor grass growth parameters and leaf $^{15}N/^{14}N$ ratio. Environmental Pollution, Volume 158(5) (2010): 1652-1660**

Effects of traffic-related nitrogenous emissions on purple moor grass (*Molinia caerulea* (L.) Moench) transplants, used here as a new biomonitoring species, were assessed along 500 m long transects orthogonal to roads located in two open areas in the Maurienne valley (French Alps). Leaves were sampled during summer 2004 and 2005 for total N-content and ^{15}N-abundance determination while nitrogen oxides (NO and NO_2) concentrations were determined using passive diffusion samplers. A significant and negative correlation was observed between plant total N-content, and ^{15}N-abundance and the logarithm of the distance to the road axis. The strongest decreases in plant N parameters were observed between 15 and 100 m from road axis. They were equivalent to background levels at a distance of about 800 m from the roads. In addition, motor vehicle pollution significantly affected vegetation at road edge, as was established from the relationship between leaf ^{15}N-abundance, total N-content and road traffic densities.

Effects of motor vehicle emissions on the $\delta^{15}N$, N-contents and growth of purple moor grass as a function of the distance to roads.

Keywords: Biomonitoring; Purple moor grass; Nitrogen oxides; ^{15}N-Abundance; Road; Traffic density; Distance

Anna KÃ¤rrman[1], JosÃ© L. Domingo[2], Xavier Llebaria[3], MartÃ Nadal[2], Esther Bigas[3], Bert van Bavel[1] and Gunilla LindstrÃm[1]. ([1]Man-Technology-Environment Research Center (MTM), School of Science and Technology, Ã–rebro University, 701 82 Ã–rebro, Sweden, [2]Laboratory of Toxicology and Environmental Health, 'Rovira i Virgili' University, Sant LlorenÃ§ 21, 43201 Reus, Catalonia, Spain, [3]Health Protection Agency, Department of Health, Generalitat de Catalunya, Roc Boronat 81-95, 08005 Barcelona, Catalonia, Spain). **Biomonitoring perfluorinated compounds in Catalonia, Spain: concentrations and trends in human liver and milk samples. Environmental Science and Pollution Research, Volume 17(3) (2010): 750-758**

Background, aim and scope

Perfluorinated compounds (PFCs) are global environmental pollutants that bioaccumulate in wildlife and humans. Laboratory experiments have revealed toxic effects such as delayed development, humoral suppression, and hepatotoxicity. Although numerous human blood levels have been reported, little is known about distribution in the human body. Knowledge about PFC distribution and accumulation in the human body is crucial to understanding uptake and subsequent effects as well as to conduct risk assessments. The present study reports PFC levels in human liver and breast milk from a general population living in Catalonia, Spain. Liver and milk levels are compared to previously reported levels in blood from the same geographic area as well as to other existing reports on human liver and milk levels in other countries.

Materials and methods

Human liver (n=12) and milk (n=10) samples were collected in 2007 and 2008 in Catalonia, Spain. Liver samples were taken postmortem from six males and six females aged 27–79 years. Milk samples were from healthy primipara women (30–39 years old). Both liver and milk were analyzed by solid-phase extraction and ultra-performance liquid chromatography tandem mass spectrometry.

Results

Six PFCs were detected in liver, with perfluorooctanesulfonate (PFOS, 26.6 ng/g wet weight) being the chemical with the highest mean concentration. Other PFCs such as perfluorohexanesulfonate (PFHxS), perfluorooctanoic acid (PFOA), and acids with chain lengths up to C11 were also detected, with mean levels ranging between 0.50 and 1.45 ng/g wet weight. On the other hand, PFOS and PFHxS were the only PFCs detected in human milk, with mean concentrations of 0.12 and 0.04 ng/mL, respectively.

Discussion

While milk concentrations were similar to reported levels from other countries, liver samples contained more PFCs above quantification limits and higher PFOS concentrations compared to the only two other reports found in the literature. Differences between the results of the present study and those concerning previous investigations can be due to declining levels of some PFCs, which have been reported for the USA. The relationship between PFC concentrations in human liver, milk, and blood was assessed using blood concentrations previously determined in Catalonia. Those levels resulted in liver/serum ratios of 1.7:1, 1.4:1, and 2.1:1 for PFOS, perfluorodecanoic acid, and perfluoroundecanoic acid, respectively. Accumulation in liver is suggested for PFOS and the perfluorocarboxylic acids with carbon chain lengths C9, C10, and C11. For PFOA and PFHxS, fivefold and 14-fold higher concentrations, respectively, were seen in serum as compared to liver. The mean concentration of PFOS and PFHxS in milk was only 0.8% and 0.6% of the reported mean serum level, respectively.

Conclusions

The results of the present study show that several PFCs could be detected in human liver samples of subjects living in Tarragona. Concerning human milk, the mechanism by which PFCs are transferred from mother's blood to breast milk is still unclear. Considering that PFCs are strongly bound to the protein fraction in blood, the possibility of PFCs entering the milk and accumulating to levels observed in maternal plasma is limited.

Recommendations and perspectives

Interestingly, the potential accumulation difference for PFCs with different chain lengths might be of great importance for risk assessment. Continuing studies on the distribution of different PFCs in human tissue are therefore justified.

Keywords: Catalonia (Spain) - LC-MS/MS - PFCs - PFOA - PFOS - Tissue distribution

Maurizio G. Paoletti[a]; Alessandra D'Incà[a]; Emanuele Tonin[a]; Stefano Tonon[a]; Carlo Migliorini[b]; Giannantonio Petruzzelli[c]; Beatrice Pezzarossa[c]; Tiziano Gomiero[a]; Daniele Sommaggio[d] . ([a] Department of Biology, Laboratory Agroecology and Ethnobiology, University of Padova, Padova, Italy, [b] VenetoAgricoltura, Settore Ricerca e Sperimentazione, Legnaro (PD), Italy, [c] *CNR*, /Istituto per lo Studio degli Ecosistemi, Sede di Pisa, Italy, [d] Dipartmtimento di Scienze e Tecnologie Agroambientali - Entomologia, Bologna University). **Soil Invertebrates as Bio-indicators in a Natural Area Converted from Agricultural Use: The Case Study of Vallevecchia-Lugugnana in North-Eastern Italy. Journal of Sustainable Agriculture, Volume 34(1) (2010): 38 - 56**

This work aims to develop a sampling methodology, based on soil invertebrates, to provide a reliable and easy-to-perform measure of environmental quality. Hand-sorting and pitfall-trapping were the main sampling systems adopted because they are quick and easy to use and do not require particular skills or tools. Both agroecosystems (organic and conventional) and seminatural environments (planted woods, hedgerows, flooded areas) have been monitored in a coastal lagoon area reclaimed to farmland in North Eastern Italy. Taxa at high hierarchical levels proved to be useful in separating different type of habitat, but were unable to provide information about the type of rural management. Carabidae (Coleoptera) seem particularly useful in studying agroecosystems: 23 species have been collected, mainly in the organic farm and in the hedgerow. The earthworm population was mainly affected by type of soil. In agroecosystems, cultivated fields had fewer individuals with respect to hedgerow, probably due to disturbance caused by soil management practices.

Keywords: invertebrate macrofauna; bioindicators; sustainability assessment; sustainable farming; ground beetles; earthworms

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Giant magnetoresistive biosensors are becoming more prevalent for sensitive, quantifiable biomolecular detection. However, in order for magnetic biosensing to become competitive with current optical protein microarray technology, there is a need to increase the number of sensors

while maintaining the high sensitivity and fast readout time characteristic of smaller arrays (1–8 sensors). In this paper, we present a circuit architecture scalable for larger sensor arrays (64 individually addressable sensors) while maintaining a high readout rate (scanning the entire array in less than 4 s). The system utilizes both time domain multiplexing and frequency domain multiplexing in order to achieve this scan rate. For the implementation, we propose a new circuit architecture that does not use a classical Wheatstone bridge to measure the small change in resistance of the sensor. Instead, an architecture designed around a transimpedance amplifier is employed. A detailed analysis of this architecture including the noise, distortion, and potential sources of errors is presented, followed by a global optimization strategy for the entire system comprising the magnetic tags, sensors, and interface electronics. To demonstrate the sensitivity, quantifiable detection of two blindly spiked samples of unknown concentrations has been performed at concentrations below the limit of detection for the enzyme-linked immunosorbent assay. Lastly, the multiplexing capability and reproducibility of the system was demonstrated by simultaneously monitoring sensors functionalized with three unique proteins at different concentrations in real-time.

Keywords: Magnetic biosensor; GMR biosensor; Spin-valve biosensor; Multiplexing spin-valves

Yufeng Han[a], Chaoling Yu[a] and Hong Liu[a]. ([a] Lab of Environmental Biology and Life Support Technology, School of Biological Science and Medical Engineering, Beihang University, Beijing 100191, China). **A microbial fuel cell as power supply for implantable medical devices. Biosensors and Bioelectronics, Volume 25(9) (2010): 2156-2160**

This study seeks a new way to provide lasting and secure power for implantable medical devices (IMDs) using a microbial fuel cell (MFC) which was proposed to be placed in human large intestine and could utilize intestinal contents and microorganisms to generate electricity. Based on the anatomic structure and inner environmental conditions of large intestine, transverse colon was chosen to be the appropriate location for the implantation of MFC. The performance of the MFC which simulated the environmental features of transverse colon by controlling dissolved oxygen (DO) and pH and was inoculated with simulated intestinal fluid (SIF) was investigated. Stable power generation of MFC was obtained after two months operation with open circuit voltage (OCV) of 552.2 mV, maximum power density of 73.3 mW/m^2, and average voltage output of 308 mV (with external resistance of 500 Ω). Moreover, the changes of environmental conditions in the chambers of MFC did not have a significant impact on human body based on the analysis of pH and DO values. Further studies on internal resistance and power density showed that the MFC could generate power of 7–10 mW according to the size of intestinal surface area, which was enough for IMDs. These results suggested that MFCs located in large intestine could be a promising power source for IMDs.

Keywords: Microbial fuel cells; Implantable medical devices; Power supply; Large intestine; Transverse colon

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Correction techniques for reproducibility and enhanced sensitivity. Biosensors and Bioelectronics, Volume 25(9) (2010): 2177-2181

Giant magnetoresistive biosensors possess great potential in biomedical applications for quantitatively detecting magnetically tagged biomolecules. Magnetic sensing does not suffer from the high background levels found in optical sensing modalities such as the enzyme linked immunosorbent assay translating into a technology with higher sensitivity. However, to reveal the full potential of these sensors and compensate for non-idealities such as temperature dependence, digital correction and calibration techniques are not only useful but imperative. Using these calibration techniques to correct for process variations and dynamic changes in the sensing environment (such as temperature and magnetic field), we are able to obtain extremely sensitive and, more importantly, reproducible results for quantifiable biomolecular reorganization. The reproducibility of the system was improved by over 3× using digital correction techniques and the sensors are made temperature independent by using a novel background correction technique.

Keywords: GMR correction algorithms; Temperature correction; Biosensors; Adaptive filter

Yong Wang[a, b], Fan Yang[a] and Xiurong Yang[a]. ([a] State Key Laboratory of Electroanalytical Chemistry, Changchun Institute of Applied Chemistry, Chinese Academy of Sciences, Changchun, Jilin 130022, China, [b] Graduate School of the Chinese Academy of Sciences, Beijing 100039, China). **Colorimetric biosensing of mercury(II) ion using unmodified gold nanoparticle probes and thrombin-binding aptamer. Biosensors and Bioelectronics, Volume 25(8) (2010): 1994-1998**

A colorimetric assay for the determination of mercury(II) (Hg^{2+}) in the presence of lead(II) (Pb^{2+}) was demonstrated with unmodified gold nanoparticles (AuNPs) as probes and 15-mer thrombin-binding aptamer (TBA, 5'-GGTTGGTGTGGTTGG-3') as sensing elements. Upon the addition of Hg^{2+} or Pb^{2+}, TBA consisting of six thymidine units and nine guanosine units interacted specifically with both ions to form a hairpin-like or a quadruplex structure, respectively. As a result, these conformation changes facilitated the salt-induced AuNP aggregation. Subsequently, to eliminate Pb^{2+} interference in the determination of Hg^{2+}, a novel technique by the use of a characteristic wavelength of aggregated AuNPs instead of the universal masking agent of Pb^{2+} (2,6-pyridinedicarboxylic acid, PDCA) was herein proposed. A comparison of the absorption spectra of the aggregated AuNPs in the presence of Hg^{2+} and Pb^{2+} showed that the characteristic wavelength of the aggregated AuNPs (800 nm) facilitated the determination of Hg^{2+} in the presence of Pb^{2+}. The calibration curve showed that the absorbance value at 800 nm increased linearly over the Hg^{2+} concentration range of 0.39–8.89 μM with a limit of detection of 200 nM. Then, the assay was successfully employed to determine Hg^{2+} in several water samples.

Keywords: Mercury(II) ion; Lead(II) ion interference; Thrombin-binding aptamer; Unmodified gold nanoparticles; Colorimetry

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Germany, [c] FORTH/IESL, Voutes Str. 1527, 71110 Heraklion, Greece, [d] Institute for Chemical Physics, Beijing Institute of Technology, Beijing 100081, People's Republic of China). Biosensor based on hydrogel optical waveguide spectroscopy. Biosensors and Bioelectronics, Volume 25(7) (2010): 1663-1668

A novel label-free biosensor based on the measurement of binding-induced refractive index changes by hydrogel optical waveguide spectroscopy (HOWS) is reported. This biosensor is implemented by using a surface plasmon resonance (SPR) optical setup in which a carboxylated poly(*N*-isoproprylacrylamide) (PNIPAAm) hydrogel film is attached on a metallic surface and modified by protein catcher molecules through amine coupling chemistry. The swollen hydrogel with micrometer thickness serves both as a binding matrix and optical waveguide. We show that compared to regular SPR biosensor with thiol self-assembled monolayer (SAM), HOWS provides an order of magnitude improved resolution in the refractive index measurements and enlarged binding capacity owing to its low damping and large swelling ratio, respectively. A model immunoassay experiment revealed that HOWS allowed detection of IgG molecules (molecular weight 150 kDa) with a 10 pM limit of detection that was 5-fold lower than that achieved for SPR with thiol SAM. For the high capacity hydrogel matrix, the affinity binding was mass transport limited. Therefore, we envisage that HOWS will provide further improved detection limit for low molecular weight analytes or for assays employing lower affinity catcher molecules.

Keywords: Optical waveguide spectroscopy; Surface plasmon resonance; Hydrogel; Label-free detection; Biosensor

Yongyan Tan[a], Xiaoxia Guo[a], Jinghui Zhang[a] and Jinqing Kan[a]. ([a] School of Chemistry and Chemical Engineering, Yangzhou University, Yangzhou 225002, PR China). Amperometric catechol biosensor based on polyaniline–polyphenol oxidase. Biosensors and Bioelectronics, Volume 25(7) (2010): 1681-1687

A novel catechol biosensor was described based on the immobilization of polyphenol oxidase (PPO) into polyaniline (PANI), which was easily constructed by direct electropolymerization of aniline in a solution containing ionic liquid, 1-ethyl-3-methylimidazolium ethyl sulfate (EMIES). The developed biosensor for the detection of catechol has a linear range of 1.25–150 μmol dm^{-3}. The maximum response current (I_{max}) and the Michaelis–Menten constant (k'_m) are 0.62 μA and 146 μmol dm^{-3}, respectively. The activation energy (E_a) of the PPO catalytic reaction is 31.1 kJ mol^{-1} in the B–R buffer. The biosensor shows good reproducibility (a relative standard deviation of 3.1% was obtained) and remarkable long-term stability (it retains 75% of the original activity after four months). The effects of potential and pH on the response current of the biosensor are also described.

Keywords: Polyphenol oxidase; Catechol; Ionic liquid; Direct electropolymerization

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Membrane proteins, which are the target of most drugs, are implicated in many critical cellular functions such as signal transduction, bioelectricity, exocytosis and endocytosis. Therefore, developing techniques to investigate the functions of membrane proteins is obviously important. Here, we have developed a novel system by integrating artificial lipid bilayer (biomimetic membrane) with single-walled carbon nanotube networks (SWNT-net) based field-effect transistor (FET), and demonstrated that such hybrid nanoelectronic biosensors can specifically and electronically detect the presence and dynamic activities of ionophores (specifically, gramicidin and calcimycin) in their native lipid environment. This technique can potentially be used to examine other membrane proteins (e.g. ligand-gated ion channels, receptors, membrane insertion toxins, and antibacterial peptides) for the purposes of biosensing, fundamental studies, or high throughput drug screening.

Keywords: Carbon nanotubes; Lipid bilayer; Bioelectronics; Membrane proteins

E. Komarova[a], K. Reber[a], M. Aldissi[a] and A. Bogomolova[a]. ([a] Fractal Systems Inc., 108 4th Street, Belleair Beach, FL 33786, USA). New multispecific array as a tool for electrochemical impedance spectroscopy-based biosensing. Biosensors and Bioelectronics, Volume 25(6) (2010): 1389-1394

Using electrochemical impedance spectroscopy (EIS) for biosensing applications typically requires repetitive experiments. To address this need, we have designed a multispecific electrochemical array with eight individually addressable 2 mm-diameter gold working electrodes for rapid biosensing data accumulation by EIS in the presence of redox agent. The array allows to incorporate multiple negative controls in the course of a single binding experiment, as well as to perform parallel identical experiments to improve reliability of detection. The array is fit with attached electrochemical cell with Ag/AgCl mini reference electrode and can be used to process macro samples of 0.5–1 ml or micro samples of 5 µl in a drop-wise fashion. Eight individual EIS measurements are completed in 15 min. The reported array is disposable, economical and is easy to use. Examples of array use for label-free genetic sensing of 2.7 kb-long target *Yersinia pestis* DNA and for protein sensing of Ricin Toxin Chain A (RTA) are presented. We suggest the reported array design as a tool for researchers in the area of EIS sensing.

Keywords: Array; Biosensor; EIS; Gold; DNA; Aptamer

Valery N. Konopsky[a] and Elena V. Alieva[a]. ([a] Institute of Spectroscopy, Russian Academy of Sciences, Troitsk, Moscow region, 142190, Russia). A biosensor based on photonic crystal surface waves with an independent registration of the liquid refractive index. Biosensors and Bioelectronics, Volume 25(5) (2010): 1212-1216

A high-precision optical biosensor technique capable of independently determining the refractive index (RI) of liquids is presented. Photonic crystal surface waves were used to detect surface binding events, while an independent registration of the critical angle was used for accurate determination of the liquid RI. This technique was tested using binding of biotin molecules to a streptavidin monolayer at low and high biotin concentrations. The attained baseline noise is 5×10^{-13} m/Hz$^{1/2}$ for adlayer thickness changes and 9×10^{-8} RIU/Hz$^{1/2}$ for RI changes.

Keywords: Label-free optical biosensors; Photonic crystal surface waves; Critical-angle refractometry; Biotin–streptavidin binding

A.M.A. Morsy[a], I.A. Ahmad[b] and A.M. Kamel[c]. ([a] Nuclear Materials Authority, P.O. Box 530, El Maadi, Cairo, Egypt, [b] Chemistry Department, Faculty of Science, Cairo University, Egypt, [c] Labeled Compounds Department, Radioisotopes Production Division, Hot Labs Center, Atomic Energy Authority, Abou-Zaabal 13759, Egypt). **Some biomedical applications of *Balanites aegyptiaca* grown naturally in radioactive area, Southeastern Desert, Egypt. Journal of Hazardous Materials, Volume 178(1-3) (2010): 725-728**

Balanites aegyptiaca is a naturally grown desert plant at some radioactive places in Wadi El-Gemal area, Southeastern Desert. The aim of the present study was to highlight on the *B. aegyptiaca* species grown naturally at radioactive places in Wadi El-Gemal area (fruit part) on the ability of using the fruit in some biomedical application (glucose, cholesterol, triglycerides, HDL-cholesterol, LDL-cholesterol and diabetes). The investigated plant was collected from different location at Wadi El-Gemal area. The uranium content was determined previously and different concentrations from the fruit with highest uranium content were used to examine the effect of *B. aegyptiaca* (fruit part) on the glucose, triglycerides, total cholesterol (HDL and LDL-cholesterol) levels using experimental rats. Different analysis techniques were used in order to determine different parameters. The obtained data suggest the beneficial role of *B. aegyptiaca* fruit as an anti-diabetic and hypo-lipidimic agent.

Keywords: *Balanites aegyptiaca*; Uranium; Glucose; Cholesterol; Triglycerides; HDL-Cholesterol; LDL-cholesterol; Diabetes

Bioengineering

Behzad Fatahi[a, b], Hadi Khabbaz[a,] and Buddhima Indraratna[c]. ([a] Faculty of Engineering, University of Technology Sydney, Broadway, Sydney, NSW 2007, Australia, [b] Coffey Geotechnics Pty Ltd., Sydney Office, Australia, [c] Faculty of Engineering, University of Wollongong, NSW 2522, Australia). **Bioengineering ground improvement considering root water uptake model. Ecological Engineering, Volume 36(2) (2010): 222-229**

Bioengineering features of native vegetation are currently being evolved to enhance soil stiffness, slope stabilisation and erosion control. The effects of tree roots on soil moisture content and ground settlement are discussed in this paper. Matric suction induced by tree roots is a key factor, governing the properties of unsaturated soils, directly imparting stability to slopes and resistance for yielding behaviour. A mathematical model for the rate of root water uptake that considers ground conditions, type of vegetation and climatic parameters has been developed. This study highlights the inter-related parameters contributing to the development of a conceptual evapo-transpiration and root moisture uptake equilibrium model that is then incorporated in a comprehensive numerical finite element model. The developed model considers fully coupled-flow-deformation behaviour of soil. Field measurements obtained by the Authors from a site in Victoria, South of Australia, are used to validate the model. In this study, the active tree root distribution has been predicted by measuring soil organic content distribution. The predicted results show acceptable agreement with the field data in spite of the assumptions made for simplifying the effects of soil heterogeneity and anisotropy. The results

prove that the proposed root water uptake model can reliably predict the region of the maximum matric suction away from the tree axis.

Keywords: Root water uptake; Native vegetation; Modelling; Soil suction; Numerical analysis

Pollen Biotechnology

Fatih Kalyoncu[1]. ([1]Faculty of Science & Arts, Department of Biology, Celal Bayar University, Manisa, Turkey). Relationship between airborne fungal allergens and meteorological factors in Manisa City, Turkey. Environmental Monitoring and Assessment, Volume 165(1-4) (2010): 553-558

In this study, the effect of relative humidity, temperature, and wind on airborne fungal allergens in the 11 different districts of Manisa City was investigated from January 2004 to December 2005. The aim of this study was to conduct a survey to get to know the relation between wind, temperature, and relative humidity and population of allergenic fungal spores in the atmosphere. A total of 792 samples were observed by using the Merck MAS100 air sampler and 12,988 fungal colonies were counted. Fourteen fungal genera could be determined; *Cladosporium* that was generally found as the predominant genus followed by *Penicillium, Aspergillus,* and *Alternaria*. During the entire study, seasonal variation was found to be related to atmospheric conditions especially. The optimal conditions of meteorological factors for the fungi growth resulted in the increased number of mycoflora, qualitatively and quantitatively.

Keywords: Fungal allergy - Temperature - Humidity - Atmosphere - Manisa - Turkey

Biotechnology Policy Issue

Daisuke Sano[a]; S. V. R. K. Prabhakar[a]. (Institute for Global Environmental Strategies, Hayama, Japan). Some Policy Suggestions for Promoting Organic Agriculture in Asia. Journal of Sustainable Agriculture, Volume 34(1) (2010):80 - 98

The benefits of organic agriculture have not been realized to their full potential, even though the organic market has been steadily expanding due to rapid economic growth and increasing environmental and health concerns in Asia. Organic agriculture has largely been promoted under the umbrella of low-chemical and environmentally friendly agriculture with different countries at different stages of establishing their own certification systems. Although overall benefits are expected from a harmonized certification system, Asian countries need to tailor their national organic promotion policies to suit diverse national circumstances and to improve their technical and institutional capacity to implement such policies.

Keywords: organic agriculture; certification; harmonization; resource conserving technologies; Community Supported Agriculture; Ecological Agriculture

Agricultural Biotechnology

De-Wei Li[1] and James LaMondia[1]. ([1]Valley Laboratory, The Connecticut Agricultural Experiment Station, 153 Cook Hill Road, Windsor, CT 06095, USA). Airborne fungi associated with ornamental plant propagation in greenhouses. Aerobiologia, Volume 26(1) (2010): 15-28

The objective was to determine potential exposure to airborne fungi in greenhouses and to characterize the temporal patterns of airborne fungi in relation to environmental conditions. We analyzed air samples collected in two greenhouses. Results showed that the top 5 fungi in greenhouse 1 were *Trichoderma*, hyphal fragments, *Aspergillus*/*Penicillium*-like, *Cladosporium*, and *Botrytis* in a descending order. Those in greenhouse 2 were *Aspergillus*/*Penicillium*-like, *Cladosporium*, *Botrytis*, yeast-like, and hyphal fragments. Maximum concentrations of *Trichoderma* and total spores in greenhouse 1 were 36,426 and 49,729 spores/m^3, respectively. Maximum concentrations of *Aspergillus*/*Penicillium* and total spores in greenhouse 2 were 46,961 and 71,037 spores/m^3, respectively. Airborne fungal populations fluctuated dramatically within 2 h during work hours, tenfold for *Aspergillus*/*Penicillium*, 66-fold for *Trichoderma*, and sevenfold for total spores. QPCR detected *Trichoderma harzianum* ranging from 7 to 3,500 conidia E/m^3. *Aspergillus*/*Penicillium* and *Botrytis* showed diurnal patterns, but not *Trichoderma*. *Aspergillus*/*Penicillium* and *Cladosporium* were positively correlated with temperature, relative humidity, dew point, heat index, and light and negatively with air movement and air pressure. *Botrytis* and *Trichoderma* were not correlated with the environmental factors. Greenhouse workers were potentially exposed up to 71,037 spores/m^3 of airborne fungi.

Keywords: Bioaerosols - Greenhouses - Occupational exposure - Propagating - Fungal spores - *Trichoderma*

Bioenergy

Miguel E Vega-Sánchez1[2] and Pamela C Ronald[1, 2]. ([1] Department of Plant Pathology, University of California, One Shields Ave., Davis, CA 95616, United States, [2] Feedstocks Division, Joint BioEnergy Institute, 5885 Hollis St., Emeryville, CA 94608, United States). Genetic and biotechnological approaches for biofuel crop improvement. Current Opinion in Biotechnology, Volume 21(2) (2010): 218-224

Research and development efforts for biofuel production are targeted at converting plant biomass into renewable liquid fuels. Major obstacles for biofuel production include lack of biofuel crop domestication, low oil yields from crop plants as well as recalcitrance of lignocellulose to chemical and enzymatic breakdown. Researchers are expanding the genetic and genomic resources available for crop improvement, elucidating lipid metabolism to facilitate manipulation of fatty acid biosynthetic pathways and studying how plant cell walls are synthesized and assembled. This knowledge will be used to produce the next generation of biofuel crops by increasing fatty acid content and by optimizing the hydrolysis of plant cell walls to release fermentable sugars.

Jyh-Ping Chen[1] and Gen-Hsu Lin[1]. ([1]Department of Chemical and Materials Engineering, Chang Gung University, Kwei-San, Taoyuan, 333, Taiwan, Republic of China). Optimization of Biodiesel Production Catalyzed by Fungus Cells Immobilized in Fibrous Supports. Applied Biochemistry and Biotechnology, Volume 161(1-8) (2010): 181-194

A circulating packed-bed bioreactor system using fibrous nonwoven fabric as the immobilization matrix was suitable for simultaneous cell growth and immobilization of *Rhizopus oryzae* fungus cells, which could be used for lipase-mediated production of biodiesel by methanolysis of soybean oil. Response surface methodology and 5-level-5-factor central composite rotatable design was proved to be a powerful tool for the optimization of methanolysis conditions catalyzed by immobilized *R. oryzae* whole cell biocatalyst. A quadratic polynomial regression model was used to analyze the relationship between the yield and the significant reaction parameters. The analysis confirmed that water content, molar ratio of methanol to oil, cell weight, and reaction time were the significant factors affecting the yield at a 95% confidence level ($p<0.05$). Under the optimum condition at 10.97% (*w*/*w*) water content, 0.64 molar ratio of methanol to oil, 2.25% (*w*/*w*) cell weight, and 23.3 h reaction time, the predicted value of yield was 72.6%. Validation experiments with yields of 70.77□±□2.46% verified the availability and the accuracy of the model.

Keywords: Biodiesel - Immobilized cells - Lipase - Response surface methodology - Nonwoven - Whole cell biocatalyst - Transesterification - Fungus cells

Fernando L. P. Pessoa[1], Shayane P. Magalhães[1] and Pedro Wagner de Carvalho Falcão[2]. ([1]DEQ—Department of Chemical Engineering, Chemistry School—Federal University of Rio de Janeiro, Av. Athos da Silveira Ramos, 149, Centro de Tecnologia, Bl. E, s. 201, Cidade Universitária, Ilha do Fundão, CEP 21941-909 Rio de Janeiro, RJ, Brazil, [2]PEQ/COPPE– Federal University of Rio de Janeiro, Av. Athos da Silveira Ramos, 149, Centro de Tecnologia, Bl. E, s. 201, Cidade Universitária, Ilha do Fundão, CEP 21941-909 Rio de Janeiro, RJ, Brazil). Production of Biodiesel via Enzymatic Ethanolysis of the Sunflower and Soybean Oils: Modeling. Applied Biochemistry and Biotechnology, Volume 161(1-8) (2010): 238-244

Biodiesel has become attractive due to its environmental benefits compared with conventional diesel. Although the enzymatic synthesis of biodiesel requires low thermal energy, low conversions of enzymatic transesterification with ethanol (ethanolysis) of oils to produce biodiesel are reported as a result of deactivation of the enzyme depending on the reaction conditions. The synthesis of biodiesel via enzymatic ethanolysis of sunflower and soybean oils was investigated. Kinetic parameters for the overall reactions were fitted to experimental data available in the literature with the Ping Pong Bi-Bi mechanism including the inhibition effect of the ethanol on the activity of lipase Novozyme® 435. The model was applied to a batch reactor and the experimental conversions were successfully reproduced. The modeling of a semibatch reactor with continuous addition of ethanol was also performed and the results showed a reduction of roughly 3 h in the reaction time in comparison with the batch-wise operation.

Keywords: Biodiesel - Enzymatic ethanolysis - Ping Pong Bi-Bi - Sunflower oil - Soybean oil

Nívea de Lima da Silva[1], Carlos Mario Garcia Santander[1], César Benedito Batistella[1], Rubens Maciel Filho[1] and Maria Regina Wolf Maciel[1]. ([1]School of Chemical Engineer, State University of Campinas (UNICAMP), P.O. Box 6066, 13081-970 Campinas, SP, Brazil). Biodiesel Production from Integration Between Reaction and Separation System: Reactive Distillation Process. Volume 161(1-8) (2010): 245-254

Biodiesel is a clean burning fuel derived from a renewable feedstock such as vegetable oil or animal fat. It is biodegradable, non-inflammable, non-toxic, and produces lesser carbon monoxide, sulfur dioxide, and unburned hydrocarbons than petroleum-based fuel. The purpose of the present work is to present an efficient process using reactive distillation columns applied to biodiesel production. Reactive distillation is the simultaneous implementation of reaction and separation within a single unit of column. Nowadays, it is appropriately called "Intensified Process". This combined operation is especially suited for the chemical reaction limited by equilibrium constraints, since one or more of the products of the reaction are continuously separated from the reactants. This work presents the biodiesel production from soybean oil and bioethanol by reactive distillation. Different variables affect the conventional biodiesel production process such as: catalyst concentration, reaction temperature, level of agitation, ethanol/soybean oil molar ratio, reaction time, and raw material type. In this study, the experimental design was used to optimize the following process variables: the catalyst concentration (from 0.5 wt.% to 1.5 wt.%), the ethanol/soybean oil molar ratio (from 3:1 to 9:1). The reactive column reflux rate was 83 ml/min, and the reaction time was 6 min.

Keywords: Biodiesel - Ethyl esters - Reactive distillation - Transesterification

Chuloo Moon[1, 2], Jae-Hyeong Ahn[1], Seung W. Kim[2], Byoung-In Sang[1] and Youngsoon Um[1]. ([1]Center for Environmental Technology Research, Korea Institute of Science and Technology, 39-1 Hawolgok-dong, Seongbuk-gu, Seoul, 136-791, South Korea, Department of Chemical and Biological Engineering, Korea University, Seoul, South Korea). Effect of Biodiesel-derived Raw Glycerol on 1,3-Propanediol Production by Different Microorganisms. Applied Biochemistry and Biotechnology, Volume 161(1-8) (2010): 502-510

The microbial production of 1,3-propanediol (1,3-PD) from raw glycerol, a byproduct of biodiesel production, is economically and environmentally advantageous. Although direct use of raw glycerol without any pretreatment is desirable, previous studies have reported that this could cause inhibition of microbial growth. In this study, we investigated the effects of raw glycerol type, different microorganisms, and pretreatment of raw glycerol on the production of 1,3-PD. Raw glycerol from waste vegetable-oil-based biodiesel production generally caused more inhibition of 1,3-PD production and microbial growth compared to raw glycerol from soybean-oil-based biodiesel production. In addition, two raw glycerol types produced from two biodiesel manufacturers using waste vegetable oil exhibited different 1,3-PD production behavior, partially due to different amounts of methanol included in the raw glycerol from the two biodiesel manufacturers. *Klebsiella* strains were generally resistant to all types of raw glycerol while the growth of *Clostridium* strains was variably inhibited depending on the type of raw glycerol. The 1,3-PD production of the *Clostridium* strains using acid-pretreated raw glycerol was significantly enhanced compared to that with raw glycerol, demonstrating the feasibility of using raw glycerol for 1,3-PD production by various microorganisms.

Keywords: Raw glycerol - 1,3-Propanediol - Fermentation - Pretreatment

Ashish Bhatnagar[1], Monica Bhatnagar[2], Senthil Chinnasamy[1] and K. C. Das[1]. ([1]Biorefining and Carbon Cycling Program, Department of Biological and Agricultural Engineering, The University of Georgia, Athens, GA 30602, USA, [2]Satellite Centre for Microalgal Biodiversity in Arid Zones of Rajasthan, Department of Microbiology, Maharshi Dayanand Saraswati University, Ajmer, 305 009, India). Chlorella minutissima—A Promising Fuel Alga for Cultivation in Municipal Wastewaters. Applied Biochemistry and Biotechnology, Volume 161(1-8) (2010): 523-536

It is imperative to slash the cost of algal oil to less than \$50 bbl^{-1} for successful algal biofuel production. Use of municipal wastewater for algal cultivation could obviate the need for freshwater and the nutrients—N and P. It would also add CO_2 through bacterial activity. *Chlorella minutissima* Fott et Nova dominated the entire phycoflora year around and through each stage of the wastewater treatment at the oxidation pond system of Wazirabad (Delhi) in India. The ability to grow so profusely in such varied and contrasting situations made this alga unique. Besides pollution tolerance, it grew heterotrophically in dark under acidic conditions and as a mixotroph in presence of light over a range of organic C substrates. It could utilize both ammoniacal and nitrate nitrogen, survived anaerobicity, 5% NaCl and −10 bar of osmotic stress. *C. minutissima* grew at pH 4–11 and raised the pH set initially by 1 to 3 units in 7.5 h. It showed gigantism and largely kept afloat in presence of utilizable organic carbon, while flocculated in mineral medium and on aging. The alga also possessed potential for biofuel production. The studied parameters indicate why *C. minutissima* was a potential biomass builder in municipal sewage and could be used to determine which other alga(e) may serve the purpose.

Keywords: Anaerobiosis - Biofuel - *Chlorella minutissima* - Mixotrophy - Wastewater

Yan Xing[1], Zhuo Li[1], Yaoting Fan[1] and Hongwei Hou[1]. ([1]Department of Chemistry, Zhengzhou University, Da Xue North Road 75, 450052 Zhengzhou, People's Republic of China). Biohydrogen production from dairy manures with acidification pretreatment by anaerobic fermentation. Environmental Science and Pollution Research, Volume 17(2) (2010): 392-399

Background, aim, and scope

Hydrogen is a clean and efficient energy source and has been deemed as one of the most promising carriers of new energy for the future. From an engineering point of view, producing hydrogen by mixed cultures is generally preferred because of lower cost, ease of control, and the possible use of organic waste as feedstock. The biological hydrogen production has been intensively studied in recent decades. So far, most investigates of biohydrogen production are still confined to using pure carbohydrates and carbohydrate-rich wastewater. Nowadays, the large amounts of livestock manure, which come from cattle feedlots, poultry, and swine buildings, are causing a major environmental issue because it has become a primary source of odors, gases, dust, and groundwater contamination. The increasingly stringent requirements for pollution control on livestock manures are challenging the scientific community to develop new waste treatment strategies. Thus, there is a pressing need to develop nonpolluting and renewable energy source utilizing the organic waste (e.g., livestock manure). It is well known that anaerobic digestion had successfully been used for the disposal of manures to produce methane in the last two decades. Recently, an alternative strategy has been developed to convert livestock

manures (e.g., dairy manures) to biohydrogen as a high value-added clean energy source instead of methane. However, little information is available on hydrogen production from dairy manure via the mixed anaerobic microbe. As far as we know, the hydrogen production is habitually accompanied with production of volatile fatty acids (VFAs), such as acetate, butyrate, and propionate, which are also an optimal feedstock for production of methane by anaerobic digestion. Provided that the biohydrogen production from dairy manure is further combined with the anaerobic digestion of the effluent from the producing hydrogen reactor that would be a one-stone two-bird paradigm, it not only produces a clean and readily usable biologic energy but also cleans up simultaneously the environment in a sustainable fashion.

Materials and methods

Prior to use, the dairy manures as natural hydrogen-producing microflora/feedstock were pretreated by infrared radiation/boiling heat by 0.2% HCl, respectively. The batch experiments were preformed with 250 mL serum vials as batch reactors filled with 100 mL mixtures, comprising the inoculum from the pre-incubated dairy manures and the feedstock from acid pretreated dairy manures as stated in Sections 2.1 and 2.2. No extra nutrients were added into the serum vials. The scale-up test was performed in a 5-L continuous stirred anaerobic bioreactor. The concentration of hydrogen, carbon dioxide, and VFAs were measured by gas chromatograph equipped with a thermal conductivity detector and a flame ionization detector, respectively. All the experiments were carried out independently in triplicates.

Results and discussion

Dairy manures with acidification pretreatment had a maximum H_2 yield of 31.5 ml/g-TVS treating 70 g/L of substrate at operating pH5.0. Meanwhile, the oxidation–reduction potential (ORP) value stayed stable at around −500 to −520 mV during the optimal hydrogen-producing period. The effluent was composed mostly of acetate and butyrate, which accounted for 78.2–81.4% of total VFAs. There was no significant methane observed in the tests. Experimental results indicated that the acidification pretreatment of dairy manure, substrate concentration, and operating pH and ORP level all had an individual significant influence on bio-H_2 production.

Conclusions

The feasibility of H_2 generation utilizing dairy manures as feedstock by anaerobic fermentation was demonstrated in this study. Biohydrogen production was found most effective utilizing acid pretreated dairy manures as feedstock at operating pH of 5.0 and substrate concentration of 70.0 g-TVS/L using pre-incubated dairy manures as inoculum. The maximal hydrogen yield of 31.5 mL H_2/g-TVS and corresponding hydrogen content of 38.6% were observed; the value was higher than previously reported.

Recommendations and perspectives

The biohydrogen production from organic wastes, such as dairy manures, is an attractive paradigm because it could produce clean biologic energy and simultaneously lean up the environment in an environmentally friendly fashion. In the present work, the biohydrogen production from dairy manures as the feedstock by mixed cultures was systematically investigated. This would provide ternary environmental benefits, viz., clean energy generation, effective method of organic waste treatment with simultaneously supplying an ideal feedstock

for methane production. It is expected that the results obtained from this work could provide some valuable information for bio-H_2 production from livestock manure.

Keywords: Batch experiments - Biohydrogen production - Dairy manure - Mixed cultures - Pretreatment - Scale-up test

Zhongfang Lei[a], Jiayi Chen[a], Zhenya Zhang[b] and Norio Sugiura[b]. ([a] Department of Environmental Science and Engineering, Fudan University, 220 Handan Road, Shanghai 200433, China, [b] Graduate School of Life and Environmental Sciences, University of Tsukuba, 1-1-1 Tennodai, Ibaraki, Tsukuba 305-8572, Japan). **Methane production from rice straw with acclimated anaerobic sludge: Effect of phosphate supplementation. Bioresource Technology, Volume 101(12) (2010): 4343-4348**

Rice straw particles were directly used as substrate for anaerobic digestion with acclimated sludge under room temperature and different levels of phosphate. Two obvious biogas production peaks were observed for all reactors, with biogas or methane yields of (0.33–0.35) m^3/kg-VS loaded or (0.27–0.29) m^3 CH_4/kg-VS loaded and average methane contents of 75.9–78.2%. A separated two-stage first-order kinetic model was developed in this study and showed a good fit to the experimental data when this complicated process was divided into two stages. The average biogas and methane production rate constants were (0.027–0.031) d^{-1} and (0.028–0.033) d^{-1}, respectively, increased by 2–3 times in the second stages than those in the first. The results indicated that an adequate level of phosphate addition (465 mg-P/L) could accelerate the biogasification process: 7–13 days earlier appearance of the two peaks and shorter time needed for complete biogasification of rice straw.

Keywords: Rice straw; Anaerobic digestion; Kinetic model; Phosphate

Stephanie Lansing[a], Jay F. Martin[b], Raúl Botero Botero[c], Tatiana Nogueira da Silva[c] and Ederson Dias da Silva[c]. ([a] Department of Environmental Science and Technology, University of Maryland, 1445 Animal Sci./Ag. Eng. Bldg., College Park, MD 20742-2315, United States, [b] Department of Food, Agricultural, and Biological Engineering, The Ohio State University, 590 Woody Hayes Drive, Columbus, OH 43210-1057, United States, [c] EARTH University, Apartado Postal 4442-1000, San Jose, Costa Rica). **Methane production in low-cost, unheated, plug-flow digesters treating swine manure and used cooking grease. Bioresource Technology, Volume 101(12) (2010): 4362-4370**

A co-digestion investigation was conducted using small-scale digesters in Costa Rica to optimize their ability to treat animal wastewater and produce renewable energy. Increases in methane production were quantified when swine manure was co-digested with used cooking grease in plug-flow digesters that operated at ambient temperate without mixing. The co-digestion experiments were conducted on 12 field-scale digesters (250 L each) using three replications of four treatment groups: the control (T0), which contained only swine manure and no waste oil, and T2.5, T5, and T10, which contained 2.5%, 5%, and 10% used cooking grease (by volume) combined with swine manure.

The T2.5 treatment had the greatest methane (CH_4) production (45 L/day), a 124% increase from the control, with a total biogas production of 67.3 L/day and 66.9% CH_4 in the produced biogas.

Increasing the grease concentration beyond T2.5 produced biogas with a lower percentage of CH_4, and thus, did not result in any additional benefits. A batch study showed that methane production could be sustained for three months in digesters that co-digested swine manure and used cooking grease without daily inputs. The investigation proved that adding small amounts of grease to the influent is a simple way to double energy production without affecting other digester benefits.

Keywords: Anaerobic digestion; Biogas; Renewable energy; Co-digestion; Waste

Victor Kraemer Wermelinger Sancho Araujo[a], Silvio Hamacher[a] and Luiz Felipe Scavarda[a]. ([a] Department of Industrial Engineering, Pontifícia Universidade Católica do Rio de Janeiro, Rua Marquês de São Vicente 225, CEP 22451-040, Rio de Janeiro, Brazil). **Economic assessment of biodiesel production from waste frying oils. Bioresource Technology, Volume 101(12) (2010): 4415-4422**

Waste frying oils (WFO) can be a good source for the production of biodiesel because this raw material is not part of the food chain, is low cost and can be used in a way that resolves environmental problems (i.e. WFO is no longer thrown into the sewage network). The goal of this article is to propose a method to evaluate the costs of biodiesel production from WFO to develop an economic assessment of this alternative. This method embraces a logistics perspective, as the cost of collection of oil from commercial producers and its delivery to biodiesel depots or plants can be relevant and is an issue that has been little explored in the academic literature. To determine the logistics cost, a mathematical programming model is proposed to solve the vehicle routing problem (VRP), which was applied in an important urban center in Brazil (Rio de Janeiro), a relevant and potential center for biodiesel production and consumption. Eighty-one biodiesel cost scenarios were compared with information on the commercialization of biodiesel in Brazil. The results obtained demonstrate the economic viability of biodiesel production from WFO in the urban center studied and the relevance of logistics in the total biodiesel production cost.

Keywords: Biodiesel; Waste frying oils; Logistics; Economic assessment; Brazil

Qun Yan[a, b], Minxing Zhao[b], Hengfeng Miao[a, b], Wenquan Ruan[a, b] and Rentao Song[c]. ([a]School of Environmental and Civil Engineering, Jiangnan University, Wuxi 214122, China, [b] Key Laboratory of Industrial Biotechnology, Ministry of Education, Jiangnan University, Wuxi 214122, China, [c] Shanghai Key Laboratory of Bio-Energy Crops, Shanghai University, Shanghai 200444, China). **Coupling of the hydrogen and polyhydroxyalkanoates (PHA) production through anaerobic digestion from Taihu blue algae. Bioresource Technology, Volume 101(12) (2010): 4508-4512**

Coupling bio-production of hydrogen and polyhydroxyalkanoates (PHA) from Taihu blue algae through metabolites circulation was investigated. It was found that the pH adjustment, especially basification was more practical and efficient than other methods for the pretreatment of blue algae before anaerobic digestion. On this occasion, SCOD, biogas accumulation and hydrogen content reached 26 mg/gTS, 500 mL and 37.2%, and which were 4.3, 1.3 and 14.4 times of those of the control group, respectively. Secondly, amounts of both butyric acid and hydrogen could be further increased when blue algae was alkali pretreated at pH 13, as the accumulation of butyric acid, acetic acid and hydrogen reached 1.7, 1.4 and 3.8 times compared to those of the control, respectively. Finally, the coupling bio-production of hydrogen and PHA was conducted through

pumping organic residues into PHA fermenter from anaerobic digester. Remarkably, it was found that the larger the pumping rate of carbon and nitrogen sources supply, the higher the yield of DCW and PHA could be expected by *Bacillus cereus*.

Keywords: Taihu blue algae; Anaerobic digestion; Coupling bio-production; Hydrogen; Polyhydroxyalkanoates (PHA)

Bor-Yann Chen[a, d], Meng-Meng Zhang[b], Chang-Tang Chang[c, d], Yongtao Ding[b], Kae-Long Lin[c, d], Chyow-San Chiou[c, d], Chung-Chuan Hsueh[a] and Huizhong Xu[b]. ([a]Department of Chemical and Materials Engineering, National I-Lan University, I-Lan 26047, Taiwan, [b] School of Environmental and Materials Engineering, Yan-Tai University, 264005, China, [c] Department of Environmental Engineering, National I-Lan University, I-Lan 26047, Taiwan, [d] Center of Green Technology (G-TEC), National I-Lan University, I-Lan 26047, Taiwan). **Assessment upon azo dye decolorization and bioelectricity generation by *Proteus hauseri*. Bioresource Technology, Volume 101(12) (2010): 4737-4741**

This study explored dye decolorization and bioelectricity generation of indigenous *Proteus hauseri* ZMd44 for dye-bearing wastewater treatment. Chemical structures of azo dyes apparently affected the performance of dye biodecolorization. Additions of diazo dye C.I. reactive blue 160 (RBu160) stimulated simultaneous dye decolorization and bioelectricity generation of ZMd44 in single chamber microbial fuel cells (MFCs). However, high-level additions of RBu160 repressed capabilities of power production in MFC due to competition of electrons used for reductive decolorization. Decolorized intermediates of RBu160-phenyl methadiamine and 5-sulfoanthranilic acid as electron shuttles might mediate electron transport for current generation in MFC.

Keywords: Azo dye decolorization; Bioelectricity generation; Microbial fuel cell; *Proteus hauseri*

Hamed M. El-Mashad[a, b] and Ruihong Zhang[b]. ([a] Department of Agricultural Engineering, Faculty of Agriculture, Mansoura University, P.O. Box 46, El-Mansoura, Egypt, [b] Biological and Agricultural Engineering Department, University of California, Davis, Davis, CA 95616, USA). **Biogas production from co-digestion of dairy manure and food waste. Bioresource Technology, Volume 101(11) (2010): 4021-4028**

The effect of manure-screening on the biogas yield of dairy manure was evaluated in batch digesters under mesophilic conditions (35 °C). Moreover, the study determined the biogas production potential of different mixtures of unscreened dairy manure and food waste and compared them with the yield from manure or food waste alone. A first-order kinetics model was developed to calculate the methane yield from different mixtures of food waste and unscreened dairy manure. The methane yields of fine and coarse fractions of screened manure and unscreened manure after 30 days were 302, 228, and 241 L/kgVS, respectively. Approximately 93%, 87%, and 90% of the biogas yields could be obtained, respectively, after 20 days of digestion. Average methane content of the biogas was 69%, 57%, and 66%, respectively. Based on mass balance calculations, separation of the coarse fraction of manure would sacrifice about 32% of the energy potential. The methane yield of the food waste was 353 L/kgVS after 30 days of digestion. Two mixtures of unscreened manure and food waste,

68/32% and 52/48%, produced methane yields of 282 and 311 L/kgVS, respectively after 30 days of digestion. After 20 days, approximately 90% and 95% of the final biogas yield could be obtained, respectively. Therefore, a hydraulic retention time (HRT) of 20 days could be recommended for a continuous digester. The average methane content was 62% and 59% for the first and second mixtures, respectively. The predicted results from the model showed that adding the food waste into a manure digester at levels up to 60% of the initial volatile solids significantly increased the methane yield for 20 days of digestion.

Keywords: Anaerobic co-digestion; Biogas; Dairy manure; Food waste; Renewable energy

Alei Geng[a, b], Yanling He[a], Changli Qian[b], Xing Yan[b] and Zhihua Zhou[b]. ([a] School of Life Science and Technology, Xi'an Jiaotong University, Xianning Rd., Xi'an 710049, PR China, [b] Key Laboratory of Synthetic Biology, Institute of Plant Physiology and Ecology, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, 300 Fenglin Rd., Shanghai 200032, PR China). **Effect of key factors on hydrogen production from cellulose in a co-culture of *Clostridium thermocellum* and *Clostridium thermopalmarium*. Bioresource Technology, Volume 101(11) (2010): 4029-4033**

A cellulolytic, hydrogen-producing bacterium (*Clostridium thermocellum* DSM 1237) and a non-cellulolytic, hydrogen-producing bacterium (*Clostridium thermopalmarium* DSM 5974) were co-cultured at 55 °C, using cellulose as the sole substrate. At a low load of cellulose (filter paper, 4.5 g/L), yeast extract had a significant effect on cellulose degradation and hydrogen production. The extent of cellulose utilization and hydrogen production displayed a linear relationship with the logarithm of the yeast extract concentration, and the optimal weight ratio of yeast extract to cellulose was 1:1. At a high load of filter paper (9 g/L), an alkali chemical was required to maintain efficient cellulose degradation. As the $KHCO_3$ concentration increased from 0 to 60 mM, the utilized cellulose increased from 1.23 g/L (13.5%) to 8.59 g/L (94.3%), and maximum hydrogen production (1387 ml/L of culture) occurred at 40 mM $KHCO_3$. Increasing the inoculation ratio of *C. thermopalmarium* to *C. thermocellum* from 0.05:1 to 0.17:1 had little influence on hydrogen production, probably because of the limited availability of soluble sugar in the medium during the early stages of the co-culture.

Keywords: Cellulose; Biohydrogen; Co-culture; Thermophilic fermentation; Real-time PCR

Yong-Zhong Wang[a], Qiang Liao[a], Xun Zhu[a], Xin Tian[a] and Chuan Zhang[a]. ([a]Institute of Engineering Thermophysics, Chongqing University, Chongqing 400030, China). **Characteristics of hydrogen production and substrate consumption of *Rhodopseudomonas palustris* CQK 01 in an immobilized-cell photobioreactor. Bioresource Technology, Volume 101(11) (2010): 4034-4041**

Effects of operation parameters on hydrogen production rate, substrate utilization efficiency and hydrogen bioconversion yield were investigated in a photobioreactor packed with sodium alginate/polyvinyl alcohol-124/carrageenan granules containing *Rhodopseudomonas palustris* CQK 01. Results demonstrate that an increase in influent glucose concentration and flow rate enhanced glucose transport from the bulk liquid into the granules, resulting in high hydrogen production. The bacteria mainly utilized the photo-heterotrophic pathway under optimal light illumination and produced hydrogen at low influent substrate loading, while they produced organic acids at high influent loading. The optimal temperature and pH of the influent medium for hydrogen production were independent of the illumination wavelength. The maximal

hydrogen production rate of 2.61 mmol/L/h was achieved under illumination at 590 nm and 6000 lx, a substrate concentration of 60 mmol/L, a temperature of 30 °C and a pH 7 of the influent medium.

Keywords: Biohydrogen production; Immobilized cells; Photobioreactor; Mass transfer; Photo-fermentation

Xiao Wu[a], Wanying Yao[b], Jun Zhu[a] and Curtis Miller[a]. ([a] Southern Research and Outreach Center, University of Minnesota, 35838 120th Street, Waseca, MN 56093, USA, [b] Department of Bioproducts and Biosystems Engineering, University of Minnesota, 1390 Eckles Avenue, St. Paul, MN 55108, USA). **Biogas and CH_4 productivity by co-digesting swine manure with three crop residues as an external carbon source. Bioresource Technology, Volume 101(11) (2010): 4042-4047**

Co-digesting swine manure with three agricultural residues, i.e., corn stalks, oat straw, and wheat straw, to enhance biogas productivity was investigated in this study. A 3 × 3 experimental design with duplicates was adopted (3 crop residues × 3 carbon/nitrogen ratios) to examine the improvement of batch digestion in terms of biogas volume produced, CH_4 content in the biogas, and net CH_4 volume. The crop residues were first cut into small sections and then ground into fine particles smaller than 40 mesh size (0.422 mm) before being added to digesters. All the digesters were run simultaneously under controlled temperature at 37 ± 0.1 °C. The length of experiment was 25 days. The results showed that all crop residues significantly increased biogas production and net CH_4 volume at all C/N ratios, among which corn stalks performed the best with increase in daily maximum biogas volume by 11.4-fold as compared to the control, followed by oat straw (8.45-fold) and wheat straw (6.12-fold) at the C/N ratio of 20/1, which was found to be the optimal C/N ratio for co-digestion in the present study. In addition, corn stalks achieved the highest CH_4 content in the biogas (~68%), which was about 11% higher than that of oat straw (~57%), whereas wheat straw and the control both had produced biogas with ~ 47% CH_4 content. Wheat straw demonstrated a lower biogas productivity than corn stalks and oat straw even it had a higher carbon content (46%) than the latter two residues (39%).

Keywords: Co-digestion; Crop residues; Swine manure; Biogas production; C/N ratios

Mohammad Pourbafrani[a, b], Gergely Forgács[a, b], Ilona Sárvári Horváth[b], Claes Niklasson[a] and Mohammad J. Taherzadeh[b]. ([a] Chemical Reaction Engineering, Chalmers University of Technology, 412 96 Göteborg, Sweden, [b] School of Engineering, University of Borås, 501 90 Borås, Sweden). **Production of biofuels, limonene and pectin from citrus wastes. Bioresource Technology, Volume 101(11) (2010): 4246-4250**

Production of ethanol, biogas, pectin and limonene from citrus wastes (CWs) by an integrated process was investigated. CWs were hydrolyzed by dilute-acid process in a pilot plant reactor equipped with an explosive drainage. Hydrolysis variables including temperature and residence time were optimized by applying a central composite rotatable experimental design (CCRD). The best sugar yield (0.41 g/g of the total dry CWs) was obtained by dilute-acid hydrolysis at 150 °C and 6 min residence time. At this condition, high solubilization of pectin present in the CWs was obtained, and 77.6% of total pectin content of CWs could be recovered by solvent recovery. Degree of esterification and ash content of produced pectin were 63.7% and 4.23%,

respectively. In addition, the limonene of the CWs was effectively removed through flashing of the hydrolyzates into an expansion tank. The sugars present in the hydrolyzates were converted to ethanol using baker's yeast, while an ethanol yield of 0.43 g/g of the fermentable sugars was obtained. Then, the stillage and the remaining solid materials of the hydrolyzed CWs were anaerobically digested to obtain biogas. In summary, one ton of CWs with 20% dry weight resulted in 39.64 l ethanol, 45 m^3 methane, 8.9 l limonene, and 38.8 kg pectin.

Keywords: Citrus waste; Ethanol; Biogas; Limonene; Pectin

Karel Komers[a], František Skopal[a] and Alexander Čegan[b]. ([a] Department of Physical Chemistry, Faculty of Chemical Technology, University of Pardubice, Czech Republic, [b] Department of Biological and Biochemical Sciences, Faculty of Chemical Technology, University of Pardubice, Czech Republic). **Continuous biodiesel production in a cascade of flow ideally stirred reactors. Bioresource Technology, Volume 101(10) (2010): 3772-3775**

The continuous methanolysis of rapeseed oil catalyzed by KOH in a cascade of 4 flow stirred reactors at a steady state of 60 °C was studied. By comparing of the determined steady state concentrations of rapeseed oil, biodiesel and KOH in the reactors (under various initial concentrations of these components and feeding) with the assumed kinetic model the rate constants of the relevant differential rate equations for rapeseed oil consumption and biodiesel production were calculated.

Keywords: Biodiesel; Production; Continuous; Flow reactors

E. Dinuccio[a], P. Balsari[a], F. Gioelli[a] and S. Menardo[a]. ([a] Department of Agriculture, Forestry, Environmental Engineering and Land Based Economics (DEIAFA), Mechanics Section – Turin University via Leonardo da Vinci 44, 10095 Grugliasco, Turin, Italy). **Evaluation of the biogas productivity potential of some Italian agro-industrial biomasses. Bioresource Technology, Volume 101(10) (2010): 3780-3783**

Batch trials were carried out to asses the biogas productivity potential of rice and barley straw, grape stalks, grape marcs, maize drying up residues, tomato skins and seeds, and whey.

Trials were carried out in 2 l glass digesters kept in a thermostate controlled room at 40 °C for 40 days. The most productive biomasses, in terms of specific methane yield, were the whey and the maize drying up residues. Their specific methane yields were 501 and 317 l_N CH_4*kg SV^{-1}, respectively. Barley and rice straw gave a specific methane yield of 229 and 195 l_N*kg VS^{-1}. Similar result was also obtained from tomato skins and seeds. Grape stalks and grape marcs produced lowest amounts of specific methane, respectively, 98 and 116 l_N CH_4*kg SV^{-1}. According to trial results and considering the availability of examined biomasses in Italy, it is possible to estimate their total energetic potential close to a value of 21,900 TJ*$year^{-1}$. This energetic potential value is equal to that obtainable from the anaerobic digestion of about 6.5 million tons of maize silage.

Keywords: Anaerobic digestion; Biogas; By-products

Senthil Chinnasamy[a], Ashish Bhatnagar[a, b], Ryan W. Hunt[a] and K.C. Das[a]. ([a]Biorefining and Carbon Cycling Program, Department of Biological and Agricultural Engineering, The University of Georgia, Athens, GA 30602, USA, [b] Arid Algae Cyanobacteria

Biodiversity and Biofuel Laboratory, Department of Microbiology, Maharshi Dayanad Saraswati University, Ajmer 305 009, India). Microalgae cultivation in a wastewater dominated by carpet mill effluents for biofuel applications. Bioresource Technology, Volume 101(9) (2010): 3097-3105

Industrial and municipal wastewaters are potential resources for production of microalgae biofuels. Dalton – the Carpet Capital of the World generates 100–115 million L of wastewater d^{-1}. A study was conducted using a wastewater containing 85–90% carpet industry effluents with 10–15% municipal sewage, to evaluate the feasibility of algal biomass and biodiesel production. Native algal strains were isolated from carpet wastewater. Preliminary growth studies indicated both fresh water and marine algae showed good growth in wastewaters. A consortium of 15 native algal isolates showed >96% nutrient removal in treated wastewater. Biomass production potential and lipid content of this consortium cultivated in treated wastewater were ~9.2–17.8 tons ha^{-1} $year^{-1}$ and 6.82%, respectively. About 63.9% of algal oil obtained from the consortium could be converted into biodiesel. However further studies on anaerobic digestion and thermochemical liquefaction are required to make this consortium approach economically viable for producing algae biofuels.

Keywords: Algae; Biodiesel; Biofuel; Carpet industry wastewater; Consortium

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The purple non-sulfur photosynthetic bacteria *Rhodopseudomonas palustris* (strain 42OL) was investigated for a co-production of both bio-H_2 and biodiesel (lipids). The investigation was carried out using malic and glutamic acids in a fed-batch cultivation system under continuous irradiances of 36, 56, 75, 151, 320, 500, and 803 W m^{-2}. Boltzmann's sigmoidal regression model was used to determine growth kinetic parameters during hydrogen photoevolution. The upper limit of volumetric hydrogen photoevolution was 15.5 ± 0.9 ml l^{-1} h^{-1}. During the entire cultivation period (408 h), the highest average hydrogen production rate (HPR_{av}) of 11.1 ± 3.1 ml l^{-1} h^{-1} was achieved at an irradiance of 320 W m^{-2}. Biomasses stored at the end of each experimental set were analyzed in order to determine lipid content, which ranged from a minimum of 22 ± 1% to a maximum of 39 ± 2% of biomass dry weight.

Keywords: Bio-H_2; Biodiesel; Hydrogen production rate; *Rhodopseudomonas palustris*; Boltzmann's sigmoidal regression model

Christos E. Papadopoulos[a], Anastasia Lazaridou[a], Asimina Koutsoumba[a], Nikolaos Kokkinos[a], Achilleas Christoforidis[b] and Nikolaos Nikolaou[a]. ([a] Kavala Institute of Technology (KIT), Department of Petroleum and Natural Gas Technology, 654 04 Ag. Loukas, Kavala, Greece, [b] Kavala Institute of Technology (KIT), Centre of Technological Research – East Macedonia and Thrace (CTR-EMTH), Ag. Loukas, Kavala, Greece).

Optimization of cotton seed biodiesel quality (critical properties) through modification of its FAME composition by highly selective homogeneous hydrogenation. Bioresource Technology, Volume 101(6) (2010): 1812-1819

The catalytic (homogeneous) hydrogenation of biodiesel's polyunsaturated fatty acid methyl esters (FAME), synthesized by transesterification of vegetable (cotton seed) oil, selectively to monounsaturated FAME, could upgrade the final quality of biodiesel. The final fuel can be optimized to have a higher cetane number and improved oxidative stability. The low-temperature performance after hydrogenation (CFPP) might be worst, but this, could be further improved through selective wintering and/or blending. The homogeneous hydrogenation of FAMEs of cotton seed biodiesel was catalyzed by the catalyst precursor $RhCl_3 \cdot 3H_20$ and STPP-TiOA. Four groups of hydrogenation experiments were carried out regarding the effects of pressure, temperature, reaction time and molecular ratio C═C/Rh. Partial hydrogenation of cotton seed FAMEs took place under mild conditions of pressure and temperature and high catalytic activities were observed in very short reaction times, and for high molecular ratios C═C/Rh.

Biodiesel's quality optimization studies, based on existing empirical models of biodiesel properties, were carried out in order to identify optimum FAME compositions and those hydrogenation conditions that could possibly supply them.

Keywords: Biodiesel properties; Empirical models; Cotton seed oil methyl esters; Partial hydrogenation; Rhodium

A. Deshpande[a], G. Anitescu[a], P.A. Rice[a] and L.L. Tavlarides[a]. ([a] Dept. of Biomedical and Chemical Engineering, Syracuse University, Syracuse, NY 13244, United States). Supercritical biodiesel production and power cogeneration: Technical and economic feasibilities. Bioresource Technology, Volume 101(6) (2010): 1834-1843

An integrated supercritical fluid technology with power cogeneration to produce biodiesel fuels, with no need for the costly separations involved with the conventional technology, is proposed, documented for technical and economic feasibility, and preliminarily designed. The core of the integrated system consists of the transesterification of various triglyceride sources (e.g., vegetable oils and animal fats) with supercritical methanol/ethanol. Part of the reaction products can be combusted by a diesel power generator integrated in the system which, in turn, provides the power needed to pressurize the system and the heat of the exhaust gases necessary in the transesterification step. The latter energy demand can also be satisfied by a fired heater, especially for higher plant capacities. Different versions of this system can be implemented based on the main target of the technology: biodiesel production or diesel engine applications, including power generation. The process options considered for biodiesel fuel production estimate break-even processing costs of biodiesel as low as \$0.26/gal (\$0.07/L) with a diesel power generator and \$0.35/gal (\$0.09/L) with a fired heater for a plant capacity of 15,000 gal/day (56,775 L/day). Both are significantly lower than the current processing costs of approximately \$0.51/gal (\$0.13/L) of biodiesel produced by conventional catalytic methods. A retail cost of biodiesel produced by the proposed method is likely to be competitive with the prices of diesel fuels.

Keywords: Biodiesel; Technical analysis; Economic assessment; Supercritical transesterification

Liaoyuan Zhang[a], Yunlong Yang[a], Jian'an Sun[a], Yaling Shen[a], Dongzhi Wei[a], Jiawen Zhu[b] and Ju Chu[c]. ([a] State Key Laboratory of Bioreactor Engineering, New World Institute of Biotechnology, East China University of Science and Technology, Shanghai 200237, PR China, [b] Chemical Engineering Research Center, East China University of Science and Technology, Shanghai 200237, PR China, [c] State Key Laboratory of Bioreactor Engineering, East China University of Science and Technology, Shanghai 200237, PR China). **Microbial production of 2,3-butanediol by a mutagenized strain of *Serratia marcescens* H30. Bioresource Technology, Volume 101(6) (2010): 1961-1967**

The production of 2,3-butanediol (2,3-BD) by *Serratia marcescens* H30 from sucrose was studied. Medium composition for 2,3-BD production by *S. marcescens* H30 was optimized in shake flask fermentations using Plackett–Burman design (PB) and response surface methodology (RSM). Results indicated that yeast extract and sodium acetate had significant effects on the 2,3-BD production. And their optimal concentrations were determined by RSM. The optimal medium was used to perform fermentation experiments by *S. marcescens* H30 in a 3.7 l bioreactor. Several feeding strategies including interim feeding, exponential feeding and constant residual sucrose concentration feeding were compared for improving 2,3-BD production. Ultimately, a suitable control strategy which combined the respiratory quotient (RQ) control with the constant residual sucrose concentration fed-batch was developed. Using this strategy, the maximum 2,3-BD concentration of 139.92 g/l with the diol (AC + BD) productivity of 3.49 g/l h and the yield of 94.67% was obtained.

Keywords: 2,3-Butanediol; *Serratia marcescens* H30; Respiratory quotient; Medium optimization; Fed-batch

Y.F. Huang[a], W.H. Kuan[b], S.L. Lo[a] and C.F. Lin[a]. ([a] Graduate Institute of Environmental Engineering, National Taiwan University, 71, Chou-Shan Road, Taipei 106, Taiwan, ROC, [b] Department of Safety, Health and Environmental Engineering, Mingchi University of Technology, 84, Gung-Juan Road, Taishan, Taipei 243, Taiwan, ROC). **Hydrogen-rich fuel gas from rice straw via microwave-induced pyrolysis. Bioresource Technology, Volume 101(6) (2010): 1968-1973**

This study aimed to research the productivity of H_2-rich fuel gas from rice straw using the microwave-induced pyrolysis. The formation constituents of gas product and the mechanism of its production were also discussed. The primary components of gas product were H_2, CO_2, CO, and CH_4, with average percentages of 50.67, 22.56, 16.09, and 7.42 vol.%, respectively. According to the TA-MS analysis, it was suggested that focused heating by microwaves made the microwave-induced pyrolysis different from the traditional pyrolysis. A chemical equation could be nearly balanced to illustrate the gas composition generated from rice straw. From the viewpoint of energy consumption, close to 60% of the input energy could be derived and utilized as bioenergy.

Keywords: Hydrogen; Fuel gas; Rice straw; Microwave-induced pyrolysis

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Chemical Engineering and Advanced Materials, Newcastle University, Newcastle upon Tyne, NE1 7RU, UK, [3]School of Earth, Atmospheric and Environmental Sciences, The University of Manchester, Manchester, UK, [4]Present address: E.ON Bioerdgas GmbH, Ruhrallee 307, 45136 Essen, Germany). The effect of flavin electron shuttles in microbial fuel cells current production. Applied Microbiology and Biotechnology, Volume 85(5) (2010): 1373-1381

The effect of electron shuttles on electron transfer to microbial fuel cell (MFC) anodes was studied in systems where direct contact with the anode was precluded. MFCs were inoculated with *Shewanella* cells, and flavins used as the electron shuttling compound. In MFCs with no added electron shuttles, flavin concentrations monitored in the MFCs' bulk liquid increased continuously with FMN as the predominant flavin. The maximum concentrations were 0.6 μM for flavin mononucleotide and 0.2 μM for riboflavin. In MFCs with added flavins, micro-molar concentrations were shown to increase current and power output. The peak current was at least four times higher in MFCs with high concentrations of flavins (4.5–5.5 μM) than in MFCs with low concentrations (0.2–0.6 μM). Although high power outputs (around 150 mW/m^2) were achieved in MFCs with high concentrations of flavins, a *Clostridium*-like bacterium along with other reactor limitations affected overall coulombic efficiencies (CE) obtained, achieving a maximum CE of 13%. Electron shuttle compounds (flavins) permitted bacteria to utilise a remote electron acceptor (anode) that was not accessible to the cells allowing current production until the electron donor (lactate) was consumed.

Keywords: Microbial fuel cell - Electron shuttle (mediator) - Flavins - *Shewanella oneidensis* MR-1 - *Clostridium*

James M. Clomburg[1] and Ramon Gonzalez[1, 2]. ([1]Department of Chemical and Biomolecular Engineering, Rice University, 6100 Main Street, MS-362, Houston, TX 77005, USA, [2]Department of Bioengineering, Rice University, Houston, TX, USA). Biofuel production in *Escherichia coli*: the role of metabolic engineering and synthetic biology. Applied Microbiology and Biotechnology, Volume 86(2) (2010): 419-434

The microbial production of biofuels is a promising avenue for the development of viable processes for the generation of fuels from sustainable resources. In order to become cost and energy effective, these processes must utilize organisms that can be optimized to efficiently produce candidate fuels from a variety of feedstocks. *Escherichia coli* has become a promising host organism for the microbial production of biofuels in part due to the ease at which this organism can be manipulated. Advancements in metabolic engineering and synthetic biology have led to the ability to efficiently engineer *E. coli* as a biocatalyst for the production of a wide variety of potential biofuels from several biomass constituents. This review focuses on recent efforts devoted to engineering *E. coli* for the production of biofuels, with emphasis on the key aspects of both the utilization of a variety of substrates as well as the synthesis of several promising biofuels. Strategies for the efficient utilization of carbohydrates, carbohydrate mixtures, and noncarbohydrate carbon sources will be discussed along with engineering efforts for the exploitation of both fermentative and nonfermentative pathways for the production of candidate biofuels such as alcohols and higher carbon biofuels derived from fatty acid and isoprenoid pathways. Continued advancements in metabolic engineering and synthetic biology will help improve not only the titers, yields, and productivities of biofuels discussed herein, but also increase the potential range of compounds that can be produced.

Keywords: Synthetic biology - Metabolic engineering - Biofuels production - *Escherichia coli* - Biofuels

Tariq Mahmood* and Syed Tajammul Hussain. (Nano Science and Catalysis Division, National Centre for Physics, Quaid-i-Azam University campus, Shahdrah valley road, Islamabad, 45320 Pakistan. *Corresponding author. E-mail: tariqm20002000@yahoo.com. Tel(office): ++92512077356. Cell: ++923335178543). **Nanobiotechnology for the production of biofuels from spent tea. African Journal of Biotechnology, Vol. 9 (6) (2010): 858-868**

Bioenergy is the only alternative and cheap source of energy which can be made easily available to the world. The present experiment included three steps for the conversion of spent tea (*Camellia sinensis*) into biofuels. In the first step, spent tea was gasified using Co nano catalyst at 300°C and atmospheric pressure. Catalytic gasification of spent tea yielded 60% liquid extract, 28% fuel gases and 12% charcoal. Gaseous products contain 53.03% ethene, 37.18% methanol and 4.59% methane. In the second step of the experiment, liquid extract of spent tea obtained from gasification, on transesterification gave 40.79% ethyl ester (biodiesel). In the third step, *Aspergillus niger's* growth on spent tea produced 57.49% bioethanol. This study reports an interesting finding that spent tea (solid waste) could be used not only for the production of biodiesel and bioethanol but also hydrocarbon fuel gases. The world today is consuming several million tons of tea yearly. The present technology could be utilized to produce alternate energy.

Keywords: Black tea, biodiesel, bioethanol, Co nano particles, hydrocarbon gases, catalytic gasification.

Narendra Mohan Verma, Shakti Mehrotra*, Amitesh Shukla and Bhartendu Nath Mishra. (Department of Biotechnology, Institute of Engineering and Technology, U.P. Technical University, Sitapur Road, Lucknow – 226021, India. *Corresponding author. E-mail: shaktimehrotra22gmail.com. narendra_106@yahoo.com. anush.elegant@gmail.com. profbnmishra @gmail.com. Tel: +91 9450713480). **Prospective of biodiesel production utilizing microalgae as the cell factories: A comprehensive discussion. African Journal of Biotechnology Vol. 9 (10) (2010): 1402-1411**

Microalgae are sunlight-driven miniature factories that convert atmospheric CO_2 to polar and neutral lipids which after esterification can be utilized as an alternative source of petroleum. Further, other metabolic products such as bioethanol and biohydrogen produced by algal cells are also being considered for the same purpose. Microaglae are more efficient than the conventional oleaginous plants in capturing solar energy as they have simpler cellular organization and high capacity to produce lipids even under nutritionally challenged and high salt concentrations. Commercially, microalgae are cultivated either in open pond systems or in closed photobioreactors. The photobioreactor systems including tubular bioreactors, plate reactors and bubble column reactors have their own advantages as they provide sterile conditions for growing algal biomass. Besides, other culture conditions such as light intensity, CO_2 concentration, nutritional balance, etc, in closed reactors remain controlled. On the other hand, though the open ponds provide a cost-effective option to utilize natural light facility for algal cells, the tough maintenance of optimal and stable growth conditions makes it difficult to manage the economy of the process. Further, these systems are much more susceptible to

contamination with unwanted microalgae and fungi, bacteria and protozoa that feed on algae. Recently, some work has been done to improve lipid production from algal biomass by implementing *in silico* and *in vitro* biochemical, genetic and metabolic engineering approaches. This article represents a comprehensive discussion about the potential of microalgae for the production of valuable lipid compounds that can be further used for biodiesel production.

Keywords: Biodiesel, fatty acid, lipids, microalgae, triacylglycerol.

Abbreviations: GHG, green house gases; DCW, dry weight biomass; TG, triacylglycerides; DOE, department of energy; ASP, aquatic species programme; *acc,* acetyl-CoA carboxilase.

Tariq Mahmood[1]*, Syed Tajammul Hussain[1] and Salman Akbar Malik[2]. ([1]Nano Science and Catalysis Division, National Centre for Physics, Quaid-i-Azam University campus, Shahdrah valley Road, Islamabad, 45320 Pakistan, [2]Department of Biochemistry, Faculty of Biological Sciences, Quaid-i-Azam University, Islamabad, 45320 Pakistan, *Corresponding author. E-mail: tariqm20002000@yahoo.com Tel: +92512077356 or +923335178543). **gene; R & D, research and development. African Journal of Biotechnology Vol. 9 (16) (2010): 2381-2391**

Three different studies were performed for the conversion of water hyacinth (*Eichhornia crassipes*) plant into biofuel. In the first study, water hyacinth was saccharified with diluted sulfuric acid (1% v/v at 110°C for one hour), fermented by yeast (*Saccharomyces cerevisiae*). The results showed the formation of 55.20% ethanol and 41.66% acetic acid. In another experiment, water hyacinth was gasified by using Ni and Co nano catalysts at 50 - 400°C and atmospheric pressure. In catalytic gasification, CH_4 (2.41 - 6.67%), C_2H_4 (19.74 - 45.52%), C_3H_4 (21.04 - 45.52%), CH_3OH (1.43 - 24.67%), and C_3H_8 / CH_3CHO (0.33 - 26.09%) products were obtained. In this study, anatase form of titanium dioxide photocatalyst was used. The reaction was performed at room temperature which gives methane, methanol and ethanol. This study also reports an interesting finding that metal contaminated water hyacinth could be used for not only the production of biofuel but also hydrocarbons.

Keywords: Water hyacinth, bioethanol, metal, cobalt nano particles, nickel nano particles, photocatalyst, biofuel, hydrocarbons.

Abbreviations: GC, Gas chromatography; **GC-MS,** gas chromatography coupled to mass spectrometry; **CID,** charge injection device; **IR,** infrared; **SEM,** scanning electron microscope; **TEM,** transmission electron microscope; **XRD,** X-ray diffraction; **HPLC,** high performance liquid chromatography; **FCC,** face-centred cubic; **PCA,** photocatalytic activity; **UV,** ultra-violet.

Rebecca M. Lennen, Drew J. Braden, Ryan M. West, James A. Dumesic, Brian F. Pfleger. (Department of Chemical and Biological Engineering, University of Wisconsin-Madison, Madison Wisconsin, telephone: 608-890-1940; fax: 608-262-5434). **A process for microbial hydrocarbon synthesis: Overproduction of fatty acids in *Escherichia coli* and catalytic conversion to alkanes. Biotechnology and Bioengineering, Volume 106(2) (2010): 193 – 202**

The development of renewable alternatives to diesel and jet fuels is highly desirable for the heavy transportation sector, and would offer benefits over the production and use of short-chain alcohols for personal transportation. Here, we report the development of a metabolically engineered strain of *Escherichia coli* that overproduces medium-chain length fatty acids via

three basic modifications: elimination of β-oxidation, overexpression of the four subunits of acetyl-CoA carboxylase, and expression of a plant acyl-acyl carrier protein (ACP) thioesterase from *Umbellularia californica* (BTE). The expression level of BTE was optimized by comparing fatty acid production from strains harboring BTE on plasmids with four different copy numbers. Expression of BTE from low copy number plasmids resulted in the highest fatty acid production. Up to a seven-fold increase in total fatty acid production was observed in engineered strains over a negative control strain (lacking β-oxidation), with a composition dominated by C_{12} and C_{14} saturated and unsaturated fatty acids. Next, a strategy for producing undecane via a combination of biotechnology and heterogeneous catalysis is demonstrated. Fatty acids were extracted from a culture of an overproducing strain into an alkane phase and fed to a Pd/C plug flow reactor, where the extracted fatty acids were decarboxylated into saturated alkanes. The result is an enriched alkane stream that can be recycled for continuous extractions. Complete conversion of C_{12} fatty acids extracted from culture to alkanes has been demonstrated yielding a concentration of 0.44 g L^{-1} (culture volume) undecane.

Keywords: metabolic engineering • *Escherichia coli* • fatty acid • alkane • biofuel • diesel

Anal Chavan[1] and Suparna Mukherji[1]. ([1]Center for Environmental Science and Engineering, Indian Institute of Technology Bombay, Powai, Mumbai, 400 076, India). **Response of an Algal Consortium to Diesel under Varying Culture Conditions. Applied Biochemistry and Biotechnology, Volume 160, Number 3(2010): 719-729**

A diesel-tolerant sessile freshwater algal consortium obtained from the vicinity of Powai Lake (Mumbai, India) was cultured in the laboratory. The presence of diesel in batch cultures enhanced the maximum specific growth rate of the algal consortium. With decrease in light–dark (L:D) cycle from 20:4 to 4:20 h, the chlorophyll-a levels decreased; however, the removal of diesel was found to be maximum at L:D of 18:6 h with 37.6% degradation over and above controls. In addition to growth in the form of green clumps, white floating biomass was found surrounding the diesel droplets on the surface. This culture predominated at the least L:D ratio of 4:20 h. Studies confirmed the ability of the floating organisms to grow heterotrophically in the dark utilizing diesel as carbon source and also in the presence of light in a medium devoid of organic carbon sources.

Keywords: Biodegradation - Cyanobacteria - Diesel - Heterotrophic growth – Hydrocarbons

Prawit Kongjan [1], Sompong O-Thong [1 2], Meher Kotay [1], Booki Min [1 3], Irini Angelidaki [1] ([*]Correspondence to Irini Angelidaki, Department of Environmental Engineering, Technical University of Denmark, DK-2800 Lyngby, Denmark; telephone: 45-4525-1429; fax: 45-4593-2850. email: Irini Angelidaki (ria@env.dtu.dk). **Biohydrogen production from wheat straw hydrolysate by dark fermentation using extreme thermophilic mixed culture Biotechnology and Bioengineering, Volume 105(5) (2010): 899 – 908**

Hydrolysate was tested as substrate for hydrogen production by extreme thermophilic mixed culture (70°C) in both batch and continuously fed reactors. Hydrogen was produced at hydrolysate concentrations up to 25% (v/v), while no hydrogen was produced at hydrolysate concentration of 30% (v/v), indicating that hydrolysate at high concentrations was inhibiting the hydrogen fermentation process. In addition, the lag phase for hydrogen production was strongly

influenced by the hydrolysate concentration, and was prolonged from approximately 11 h at the hydrolysate concentrations below 20% (v/v) to 38 h at the hydrolysate concentration of 25% (v/v). The maximum hydrogen yield as determined in batch assays was 318.4 ± 5.2 mL-H_2/g-sugars (14.2 ± 0.2 mmol-H_2/g-sugars) at the hydrolysate concentration of 5% (v/v). Continuously fed, and the continuously stirred tank reactor (CSTR), operating at 3 day hydraulic retention time (HRT) and fed with 20% (v/v) hydrolysate could successfully produce hydrogen. The hydrogen yield and production rate were 178.0 ± 10.1 mL-H_2/g-sugars (7.9 ± 0.4 mmol H_2/g-sugars) and 184.0 ± 10.7 mL-H_2/day $L_{reactor}$ (8.2 ± 0.5 mmol-H_2/day $L_{reactor}$), respectively, corresponding to 12% of the chemical oxygen demand (COD) from sugars. Additionally, it was found that toxic compounds, furfural and hydroxymethylfurfural (HMF), contained in the hydrolysate were effectively degraded in the CSTR, and their concentrations were reduced from 50 and 28 mg/L, respectively, to undetectable concentrations in the effluent. Phylogenetic analysis of the mixed culture revealed that members involved hydrogen producers in both batch and CSTR reactors were phylogenetically related to the *Caldanaerobacter subteraneus*, *Thermoanaerobacter subteraneus*, and *Thermoanaerobacterium thermosaccharolyticum*.

Keywords: hemicelluloses • biohydrogen • mixed culture fermentation • microbial community dynamics • extreme thermophilic conditions

Name of Journals

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2. Aerobiologia
3. Annual Review-Plant Pathology
4. Annual Review- Ecology and Systematics
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7. Annual Review-Biophysics and Biomolecular Structure
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