

EMCB-ENVIS Node
on

ENVIRONMENTAL BIOTECHNOLOGY

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C O N T E N T S

Title	Page No.
1. Background	4
2. Abstract format	5
3. General information	6
4. Abbreviation used	9
5. Abstracts	
Bioaccumulation	12
Biodegradation	30
Bioenergy	66
Bioengineering	73
Biofertilizer	75
Biomarker	75
Biopesticide	77
Bioremediation	80
Biosensor	91
Biotechnology Policy Issue	95
Biotransformation	98
6. Author Index	107
7. Name of Journal	117
8. List of Libraries	118

BACKGROUND

Environmental Information System (ENVIS) is established in the year 1984 as a network of Information Centre. It is planned by the Ministry of Environment and Forest. Aim of this centre is to provide descriptive data and environmental subject related numerical data. Now 35 centres are working under this network on various subjects area in the country. The focal point of this network is at the Ministry of Environmental and Forest, Government of India, New Delhi.

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

The objective of this centre is to collect data, related to the above mentioned subject, from different major libraries in Kolkata with different journals, Annual reviews, Internet and to generate a database and to create a website with this database. View point of this journal abstract is to help the interested research workers, scientist, administrator and the public.

This is the fourth publication of this ENVIS Centre. This contains the abstract of research papers collected in the area of Environmental Biotechnology from various journal published during December 2000 onwards. Here various topics like Bio-engineering, Bio-degradation, Bio-remediation, Bio-transformation etc. are covered. We are grateful to the various libraries and their staff for their extended cooperation in the collection of the articles.

Abstract Format

The format of the abstract is as follows:

Abstract : The abstracts were arranged in alphabetic orders 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 author name. When the address of any other author is found, it is written after wards delimited by stop(.).

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

GENERAL INFORMATION

Abstract have been taken directly from source document like research report, journals, Internet, seminars, proceedings, standards and patents. All the resources published within the year 2000-2003.

Abstract are broadly classified and arranged under the following heads:

Bioaccumulation: It studies address the buildup of bioaccumulative compounds through biomagnification and/or bioconcentration. 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 any time they are taken up and stored 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. These 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.

Because the microorganisms already occur naturally in the environment they pose no contamination risk.

Bio-Transformation: This is a process of Biological changes of complex compound to simpler toxic to non-toxic or vice-versa. Several microorganism are capable of transforming a variety of compound founding nature but generally with respect to synthetic compound they are unable to show any appropriate action. Biotransfer appears to be one of the major detoxication method known so far.

Biomarker: It is a biological response to a chemical that gives a measure of exposure and, sometimes, of toxic effect. Biological markers found in crude oils and source rock

extracts can provide molecular evidence of the correlation among oils and their sources.

Bioenergy: In recent decades, efforts were made for evolving were non-polluting bioenergy sources or energy generation from organic waste or biomass. These are all ecofriendly solution. Biomass energy supply demand balances have become a component of energy sector analysis and planning and assumed greater importance in countries. These are variety of biological energy sources. Biomass, Biogas, Hydrogen are the example of Bioenergy.

Biofertilizer: To reduce the impact of excess chemical fertilizers in the field of agriculture the biofertilizer is 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 were 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 involves combining organic materials under conditions that enables them to decompose more quickly than they would in nature.

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

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

In nature, there is no 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 will 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 will detect the presence and measure the quantities of specific substances in a variety of environments. These specific substances may include sugars, proteins, or humas and variety of toxins in the industrial effluents. In designing a biosensor an enzyme or an antibody or even microbial cells are associated with microchip devices which are used for quantitative estimate of a substance.

Bioengineering: It is a developing speciality featuring a multidisciplinary approach to the solution of problems in medicine and biology, based on the application of advances in science, engineering and technology. A major focus for bioengineering is to improve the quality of life of people with medical conditions that restrict independent living and integration within the community.

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

Biotechnology Policy Issue: Biotechnology appears to be a 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 was made in diverse field of agriculture by applying Biotechnology. It includes development of genetically modified crops, genetically improvement in sericulture practices, improvement in Biofertilizer development and similar other aspects. Crop production against pest and disease stress resistance of crops also considered to be emerging area of Agricultural Biotechnology.

ABBREVIATIONS USED IN ADDRESSES AND CITED JOURNALS

Acad	Academy	Chem	Chemistry
Adm	Administration	Chemi	Chemical
Admn	Administrative	Clini	Clinical
Adv	Advance	Co	Company
Agric	Agriculture	Coil	College
Agrici	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	Conti	Control
Appt	Applied	Contam	Contamination
Arch	Archives	Corp	Corporation
Archaeo	Archaeology	Coun	Council
Archaeol	Archaeological	Cult	Culture
Architect	Architecture	Cultl	Cultural
Assoc	Association	Curr	Current
Asst	Assistant	Dept	Department
Atom	Atomic	Dev	Development
Bacterio	Bacteriology	Develop	Developmental
Bacteriol	Bacteriological	Dig	Digest
Bd	Board	Div	Division
Bio	Biology	Divl	Divisional
Biochem	Biochemistry	Dte	Directorate
Biochemi	Biochemical	Dy	Deputy
Bioengng	Bioengineering	Eco	Ecology
Biol	Biological	Ecol	Ecological
Biometeo	Biometeorology	Econ	Economics
Biophys	Biophysics	Ecosys	Ecosystem

Biometeol	Biometeorological	Exotoxico	Ecotoxicology
Biotech	Biotechnique(s)	Endocrinol	Endocrinologica l
Biotechno	Biotechnology	Engng	Engineering
Biotechnol	Bitechnological	Engrs	Engineers
Bidg	Building	Env	Environment
Bot	Botany	Environ	Environmental
Boti	Botanical	Epidemic	Epidemiology
Br	Branch	Epidemiol	Epidemiological
Bull	Bulletin	Estb	Establishment
Cent	Centre	Ethnopharma co	Ethnopharmaco logy
Centl	Central	Exot	Experiment

Expti	Experimental	Microbiol	Microbiologica l
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
Geogri	Geographical	Occ	Occassional
Geol	Geological	Occupl	Occupational
Geosci	Geoscience	Oceanogr	Oceanogoraph y
Govt	Government	Org	Organic
Hist	History	Orgn	Organisation
Hlth	Health	Pharmaco	Pharmacology
Hort	Horticulture	Pharmacol	Pharmacologic al
Hosp	Hospital	Phyl	Physical
Hydro	Hydrology	Patho	Pathology
Hydrol	Hydrological	Pathol	Pathological
Immuno	Immunology	Petrochemi	Petrochemical
Immunol	Immunological	Petro	Petrology
Ind	Industry	PG	Post Graduate
Inf	Information	Phys	Physics
Inst	Institute	Physio	Physiology
Instn	Institution	Phytopath	Phytopatholog y
Int	International	Phytopathol	Phytopatholog ical
Irrig	Irrigation	Plang	Planning
J	Journal	Polln	Pollution

Lab	Laboratory	Proc	Proceedings
Lett	Letter(s)	Prot	Protection
Ltd	Limited	Pub	Publication
Malario	Malariology	Pvt	Private
Malariol	Malariological	Qlty	Quality
Manag	Management	Qr	Quarter
Med	Medicine	Rad	Radiation
Medl	Medical	Radio	Radiology
Metab	Metabolism	Radiol	Radiological
Metall	Metallurgy	Rd	Road
Metallurg	Metallurgical	Recd	Received
Meteo	Meteorology	Reg	Region
Meteol	Meteorological	Regl	Regional
Microbio	Microbiology		

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

Bioaccumulation

A. Binelli, A. Provini. (Department of Biology, University of Milan, Via Celoria 26, 20133, Milan, Italy). The PCB pollution of Lake Iseo (N. Italy) and the role of biomagnification in the pelagic food web. *Chemosphere*, 53(2) (2003), 143-151.

Several models of varying complexity have been used to predict pollutant concentrations in the higher levels of the food web from those in lower levels, but the role of the biomagnification process in aquatic food chains is still controversial. We used the fugacity-based approach to verify the transfer of PCBs through the pelagic food chain of Lake Iseo (N. Italy), sampling several zebra mussel specimens and some fish belonging of different trophic levels. The zebra mussel seems to be a suitable starting species for modelling the bioaccumulation process through the trophic web, not only because its physiological characteristics and population size do not change much with time (as do algae and zooplankton) but also because it takes up toxicants exclusively from the water, as shown by the application of two predictive trophic models commonly used. The data provided by one of those models were in good agreement with our experimental data on fish in Lake Iseo, that show a not negligible uptake from food for the top predator species (pike and perch) with an increase of about three times in comparison with the PCB levels measured in the zebra mussel specimens.

Andrès Y, Texier A.C., Le Cloirec P. Rare Earth Elements Removal By Microbial Biosorption: A Review. *Environmental Technology*, 24(11) (2003), 1367-1375.

This paper reviews published work on the sorption of rare earth elements by microbial biomass. In a first part, the biosorption capacities and the various experimental conditions performed in batch reactor experiments are compared. Secondly, sorption modelling generally used in biosorption studies are described. Thirdly, the microbial cell wall characteristics of the metallic ion binding sites are considered. From these observations it seems that the important functional groups for metallic ion fixation are the carboxyl and the phosphate moieties. Moreover, the competing effect of various ions like aluminium, iron, glutamate, sulphate etc. is described. Finally, some adsorption results of the rare earth elements in dynamic reactors are presented.

Bénédicte Viard, François Pihan, Sandrine Promeyrat, Jean-Claude Pihan. (Laboratoire BFE--Equipe PEE, Université de Metz, Campus Bridoux, 2 rue du général Delestraint, 57070, Metz, France). Integrated assessment of heavy metal (Pb, Zn, Cd) highway pollution: bioaccumulation in soil, Graminaceae and land snails. *Chemosphere*, 55(10) (2004), 1349-1359.

To assess the contamination induced by traffic at the vicinity of a highway (A31, France), several complementary studies were carried out on two sites, with different profiles and traffic intensity. Concentrations of zinc, lead and cadmium were measured by atomic absorption spectrophotometry in deposits, roadside soil and autochthonous plants (Graminaceae) gathered at the vicinity of the highway (1-320 m), and in the viscera of snails *Helix aspersa*, transferred as sentinel in the sites. According to the results obtained for different compartments, the highway induces a contamination on the surrounding environment, up to 320 m, but with the maximum contamination observed between 5 and 20 m: the concentrations measured in plants at the vicinity of the highway were 2.1 mg Pb kg⁻¹ DW, 0.06 mg Cd kg⁻¹ DW, 62 mg Zn kg⁻¹ DW and the concentrations measured in snails were 21.3 mg Pb kg⁻¹ DW, 5.7 mg Cd kg⁻¹ DW, 510.8 mg Zn kg⁻¹ DW.

The levels measured decreased with increasing distance from the highway. Results of the three metals studied indicated that lead seems to be the best metal to evaluate road transport contamination.

Bernard Lachance, Agnès Y. Renoux, Manon Sarrazin, Jalal Hawari and Geoffrey I. Sunahara. (Applied Ecotoxicology Group, Biotechnology Research Institute, National Research Council of Canada, 6100 Royalmount Ave., Montreal, QC, Canada H4P 2R2). Toxicity and bioaccumulation of reduced TNT metabolites in the earthworm *Eisenia andrei* exposed to amended forest soil. *Chemosphere*, 55(10) (2004), 1339-1348.

Soils contaminated with 2,4,6-trinitrotoluene (TNT) and TNT primary reduction products have been found to be toxic to certain soil invertebrates, such as earthworms. The mechanism of toxicity of TNT and of its by-products is still not known. To ascertain if one of the TNT reduction products underlies TNT toxicity, we tested the toxicity and bioaccumulation of TNT reduction products. 2-Amino-4,6-dinitrotoluene (2-ADNT), 4-amino-2,6-dinitrotoluene (4-ADNT), 2,4-diamino-6-nitrotoluene (2,4-DANT) and 2,6-diamino-4-nitrotoluene (2,6-DANT) were tested separately in adult earthworms (*Eisenia andrei*) following a 14-d exposure to amended sandy loam forest soil. TNT, 4-ADNT, and 2-ADNT were lethal to earthworms (14-d LC50 were: 580, 531 and 1088 $\mu\text{mol kg}^{-1}$, or 132, 105 and 215 mg kg^{-1} dry soil, respectively) and gave the following order of toxicity: 4-ADNT > TNT > 2-ADNT. Exposure to 2,4-DANT and to 2,6-DANT caused no mortality at 600 $\mu\text{mol kg}^{-1}$ or 100 mg kg^{-1} dry soil. We found that all four TNT reduction products accumulated in earthworm tissues and 2-ADNT reached the highest levels at $3.0 \pm 0.3 \mu\text{mol g}^{-1}$ tissue. The 14-d bioaccumulation factors were 5.1, 6.4, 5.1 and 3.2 for 2-ADNT, 4-ADNT, 2,4-DANT and 2,6-DANT, respectively. Results also suggest that some TNT metabolites are at least as toxic as TNT and should be considered when evaluating the overall toxicity of TNT-contaminated soil to earthworms.

César Ricardo Teixeira Tarley, Marco Aurélio Zezzi Arruda. (Department of Analytical Chemistry, Institute of Chemistry, State University of Campinas--UNICAMP, P.O. Box 6154, Campinas 13084-971, São Paulo, Brazil). Biosorption of heavy metals using rice milling by-products. Characterisation and application for removal of metals from aqueous effluents. *Chemosphere*, 54(7) (2004), 987-995.

The morphological characteristics as well as chemical composition of rice husks were evaluated by different techniques such as spectroscopy and thermogravimetry. The material, which is considered a by-product obtained from rice milling, was then investigated as a potential decontaminant of toxic heavy metals present in laboratory effluents. Studies using glass columns were carried out at room temperature employing 100 ml of synthetic solutions containing Cd(II) and Pb(II) at 100 mg l^{-1} in order to study the effects of pH, flow rate and particle size on Cd(II) and Pb(II) adsorption. After establishing the optimised conditions, the potentiality of rice husks for removing Cd(II) and Pb(II) ions from 100 ml of laboratory effluent, presenting concentrations before treatment of 22 and 12 mg l^{-1} , respectively, was evaluated. The ability to take up other metals species, such as Al(III), Cu(II) and Zn(II), present in this effluent was also studied. According to the data obtained, under the optimised conditions (pH=4.0, flow rate of 8.0 ml min^{-1} and $\leq 355 \mu\text{m}$ rice husk particle size), 30 g of husks were necessary to attain the permissible limits for effluent release, as recommend by the EPA, for those species evolved in this work (Al, Cd, Cu, Pb and Zn).

Chaisuksant Y. Biosorption of Cadmium (II) and Copper (II) by Pretreated Biomass of Marine Alga *Gracilaria Fisheri*. *Environmental Technology*, 24(12) (2003), 1501-1508.

The cadmium (II) and copper (II) adsorption properties of chemically pretreated biomass of red marine alga *Gracilaria fisheri* were investigated. Batch equilibrium experiments showed that the maximum adsorption capacity values of the pretreated biomass for cadmium and copper were 0.63 and 0.72 mmol g⁻¹, respectively. The equilibrium data fitted well to the Langmuir isotherm model. The adsorption capacity increased as pH increased and reached a plateau at pH 4.0. The cadmium and copper uptake rates were rapid with 90% of the biosorption completed within 30 minutes. The presence of light metal ions (Na⁺, K⁺, Mg²⁺ and Ca²⁺) in solution had an insignificant effect on cadmium and copper sorption capacity. These findings indicate a positive potential for the biosorbent development with effective heavy metal removal capacity in the presence of light metal ions in waste streams by using the biomass of plentifully available red marine algae.

Chang-Tang Chang, Bor-Yann Chen, I-Shing Shiu, Fu-Teng Jeng. (Department of Environmental Engineering, National I-Lan University, I-Lan, Taiwan. Department of Chemical Engineering, National I-Lan University, I-Lan, Taiwan. Graduate Institute of Environmental Engineering, National Taiwan University, Taipei, Taiwan). Biofiltration of trimethylamine-containing waste gas by entrapped mixed microbial cells. *Chemosphere*, 55(5) (2004), 751-756.

The study provides novel attempt to use an aerobic biofiltration system containing entrapped mixed microbial cells (EMMC) for removal of (CH₃)₃N-dominant waste gases. In the study, heterotrophic microflora-immobilized cellulose was packed into an EMMC reactor to degrade (CH₃)₃N. Effects of (CH₃)₃N inlet concentrations in continuous mode of operation at various flow rates are indicated. The result indicated that the (CH₃)₃N removal efficiency is higher than 90% at inlet loading below 27.2 mg N h⁻¹ and retention time 5.3 min. In addition, the maximal mass loading to reach approximately 99% efficiency was 95.5 mg N h⁻¹ for trimethylamine treatment. This EMMC biofiltration system also showed higher tolerance to endure fluctuations in concentrations and flow rates and still maintained in stable performance for removal. Adaptability test in response to gradual shift up and down of inlet TMA loading indicated that lack of steady-state multiplicity and hysteresis guarantees the microbial communities more precisely adapted to continuous treatment for maintaining stability.

Costantino Vischetti, Ettore Capri, Marco Trevisan, Cristiano Casucci, Piero Perucci. (Dipartimento di Scienze Ambientali e delle Produzioni Vegetali, Università Politecnica delle Marche, via Brecce Bianche, 60131, Ancona, Italy. Istituto di Chimica Agraria ed Ambientale, Università Cattolica del Sacro Cuore, via Emilia Parmense, Piacenza, Italy). Biomassbed: a biological system to reduce pesticide point contamination at farm level. *Chemosphere*, 55(6) (2004), 823-828.

A potential method for cleaning water from point-source pollution by organic compounds is using biological reactors. In this study, four reactors were tested for their ability to retain and degrade pesticides. The pesticides tested were the insecticide chlorpyrifos, the fungicide metalaxyl and the herbicide imazamox. The reactors were filled with differing mixtures of vine-branch, citrus peel, urban waste and public green compost. The reactor volume was 188 l. Forced circulation of the contaminated solution was programmed to decontaminate the solution. Both retention and degradation of the compounds by the reactors was studied. Chlorpyrifos was the best retained, due to its physico-chemical characteristics, while only one substrate effectively retained metalaxyl and imazamox (citrus peel + urban waste compost). Degradation of the pesticides in the reactors was faster than published values for degradation in soil. The half-life of all pesticides in the reactors was less than 14 days, compared to literature values of 60–70 days in soil. The combined retention and fast degradation make the biofilter a feasible technique to reduce spill-related and point environmental contamination by pesticides. The technique is most effective against

persistent pesticides, while for mobile pesticides, the efficiency can be improved with several passages of the contaminated solution through biofilters.

D. G. Heijerick, C. R. Janssen, C. Karlèn, I. Odnevall Wallinder, C. Leygraf. (Laboratory of Environmental Toxicology and Aquatic Ecology, Ghent University, J. Plateaustraat 22, B-9000 Ghent, Belgium. Department of Materials Science and Engineering, Division of Corrosion Science, Royal Institute of Technology, SE-100 44 Stockholm, Sweden). Bioavailability of zinc in runoff water from roofing materials. *Chemosphere*, 47(10) (2002), 1073-1080.

Corrosion and runoff from zinc-coated materials and outdoor structures is an important source for the dispersion of zinc in the environment. Being part of a large inter-disciplinary research project, this study presents the bioavailability of zinc in runoff water immediately after release from the surface of 15 different commercially available zinc-based materials exposed to the urban environment of Stockholm, Sweden. Runoff water was analysed chemically and evaluated for its possible environmental impact, using both a biosensor test with the bacteria *Alcaligenes eutrophus* (Biomet®) and the conventional 72 h growth inhibition test with the green alga *Raphidocelis subcapitata*. Chemical speciation modelling revealed that most zinc (94.3–99.9%) was present as the free Zn ion, the most bioavailable speciation form. These findings were confirmed by the results of the biosensor test (Biomet®) which indicated that all zinc was indeed bioavailable. Analysis of the ecotoxicity data also suggested that the observed toxic effects were due to the presence of Zn²⁺ ions. Finally, regression analysis showed that, for this type of runoff samples, the rapid screening biosensor was capable of predicting (a) the total amount of zinc present in the runoff samples (R^2 of 0.93–0.98; $p < 0.05$) and (b) the observed 72 h-EbC_{50s} (R^2 of 0.69–0.97; $p < 0.05$).

Epaminondas Voutsas, Kostis Magoulas, Dimitrios Tassios. (Thermodynamics and Transport Phenomena Laboratory, Department of Chemical Engineering, Section II, National Technical University of Athens, 9, Heron Polytechniou Street, Zografos GR-15780, Athens, Greece). Prediction of the bioaccumulation of persistent organic pollutants in aquatic food webs. *Chemosphere*, 48(7) (2002), 645-651.

Predictive correlations of the bioaccumulation factor of persistent organic pollutants in aquatic biota are presented as functions of their octanol/water partition coefficient. The correlations demonstrate the importance of differentiating among the different levels in the food web and of accounting for the pollutant's bioavailability by considering the amount freely dissolved in water instead of the total concentration. They also reveal the significance of the pollutant's octanol/water partition coefficient value on its biomagnification along the levels of the trophic chain. Prediction results, finally, demonstrate that the correlations provide reasonably accurate estimates of bioaccumulation, typically within an order-of-magnitude.

Gerhardt F. Riedel, Nathalie Valette-Silver. (Academy of Natural Sciences, Estuarine Research Center, 10545 Mackall Road, St. Leonard, MD 20685, USA. National Oceanic and Atmospheric Administration, National Ocean Service, 1305 East West Highway, Silver Spring, MD 20910, USA). Differences in the bioaccumulation of arsenic by oysters from Southeast coastal US and Chesapeake Bay: environmental versus genetic control. *Chemosphere*, 49(1) (2002), 27-37.

The potential sources of relatively great concentrations of arsenic (As) in oysters from the Southeastern United States coast was examined in a study conducted from August 1998 through October 1999. A transplant experiment was conducted to determine whether genetic or environmental differences accounted for the observed difference between

Southeastern oysters, and oysters elsewhere on the east coast. Oysters originating in South Carolina (a region where As in oysters is usually greater) and Maryland (a region where arsenic in oysters is less) were reciprocally transplanted to determine whether site of growth or site of origin would determine the accumulation of As. To examine the potential role of various potential sources of As exposure on the concentrations of As in oysters, samples of native oysters, water, pore water and suspended particles were collected and analyzed for As monthly, while the sediments were examined four times during the year. Concentrations of As in transplanted oysters matched the concentrations of As in oysters native to the area in which they were grown, rather than that of oysters from their site of origin. Oysters from South Carolina had average concentrations of As \approx 3.2 times that of oysters from Maryland. This enrichment was similar to enrichments of water (3.4 times), sediment (2.5 times), suspended particles (1.7 times), and pore water (3.1 times) from South Carolina compared to Maryland. This supports the hypothesis that the cause of the apparent As enrichments in the Southeastern oysters is environmental, but leaves the question of the primary source for arsenic incorporation by oysters open.

Hai Yan, Gang Pan. (State Key Laboratory of Environmental Aquatic Chemistry, Research Center for Eco-environmental Sciences, Chinese Academy of Sciences, P.O. Box 18, Beijing 100085, China. Eco-environmental Chemistry Laboratory, Qingdao Institute of Chemical Technology, Qingdao 266042, China). Increase in biodegradation of dimethyl phthalate by *Closterium lunula* using inorganic carbon. *Chemosphere*, 55(9) (2004), 1281-1285.

The effect and mechanism of inorganic carbon (IC) on the biodegradation of dimethyl phthalate (DMP) by a green microalga *Closterium lunula* was investigated. The growth of this microalga and the biodegradation of DMP were significantly enhanced when the initial IC was increased. An intermediate product of DMP biodegradation was identified as phthalic acid (PA) that was accumulated and caused a sharp decrease in pH of microalgal culture medium, which inhibited both the growth of microalga and the biodegradation of DMP. A suggested second-order kinetic equation of organic pollutant biodegradation by microalgae ($-dC/dt=kNr$) fitted well with the experimental data. The increase of IC caused a decline in biodegradation rate constant for organic carbon (k) and an increase in growth (N) by supplying a favorite carbon source and mitigating the decrease of pH. As the net effect, the overall biodegradation rate of DMP was promoted as IC increased, which was dominated by the increase of microalgal growth.

Hai Yan, Gang Pan. (State Key Laboratory of Environmental Aquatic Chemistry, Research Center for Eco-environmental Sciences, Chinese Academy of Sciences, P.O. Box 2871, Beijing 100085, China. Eco-environmental Chemistry Laboratory, Qingdao Institute of Chemical Technology, Qingdao 266042, China). Toxicity and bioaccumulation of copper in three green microalgal species. *Chemosphere*, 49(5) (2002), 471-476.

The effective concentrations of copper on the inhibition of the growth of *Scenedesmus obliquus*, *Chlorella pyrenoidosa* and *Closterium lunula* at 96 h (96 h EC₅₀) were determined to be 50, 68 and 200 $\mu\text{g/l}$, respectively. The low initial bioaccumulation of Cu by *C. lunula* was found to be responsible for its tolerance to Cu. The amount of Cu accumulated by all three microalgae reached the maximum value and decreased quickly after the peak followed by a slow decrease over the next 6 d. Bioaccumulation of Cu by *C. lunula* was directly proportional to the initial Cu concentration. After reaching the first peak after 1 d, the bioconcentration factor of Cu by microalgae declined to its minimum value during the exponential growth phase but increased in the stationary growth phase again. This indicates that desorption of Cu from microalgae was higher during the exponential growth phase but

lower in the stationary growth phase. Smaller microalgae with low 96 h EC50 values are more efficient in removing Cu from wastewater.

Hangzhou. (Institute of Food and Agricultural Science, University of Florida, Indian River Research and Education Center, 2199 South Rock Road, Fort Pierce, FL 34945-3138, USA). Growth response and phytoextraction of copper at different levels in soils by *Elsholtzia splendens*. *Chemosphere*, 55(9) (2004), 1179-1187.

Phytoremediation is a promising approach for cleaning up soils contaminated with heavy metals. Information is needed to understand growth response and uptake mechanisms of heavy metals by some plant species with exceptional capability in absorbing and superaccumulating metals from soils. Greenhouse study, field trial, and old mined area survey were conducted to evaluate growth response and Cu phytoextraction of *Elsholtzia splendens* in contaminated soils, which has been recently identified to be tolerant to high Cu concentration and have great potential in remediating contaminated soils. The results from this study indicate that the plant exhibited high tolerance to Cu toxicity in the soils, and normal growth was attained up to 80 mg kg⁻¹ available soil Cu (the NH₄OAc extractable Cu) or 1000 mg kg⁻¹ total Cu. Under the field conditions, a biomass yield of 9 ton ha⁻¹ was recorded at the soil available Cu level of 77 mg kg⁻¹, as estimated by the NH₄OAc extraction method. Concentration-dependent uptake of Cu by the plant occurred mainly at the early growth stage, and at the late stage, there is no difference in shoot Cu concentrations grown at different extractable soil Cu levels. The extractability of Cu from the highly polluted soil is much greater by the roots than that by the shoots. The NH₄OAc extractable Cu level in the polluted soil was reduced from 78 to 55 mg kg⁻¹ in the soil after phytoextraction and removal of Cu by the plant species for one growth season. The depletion of extractable Cu level in the rhizosphere was noted grown in the mined area, even at high Cu levels, the NH₄OAc extractable Cu in the rhizosphere was 30% lower than that in the bulk soil. These results indicate that phytoextraction of *E. splendens* can effectively reduce the plant-available Cu level in the polluted soils.

J. A. Fernández, A. Carballeira. (Ecología, Facultad de Biología, Universidad de Santiago de Compostela, 15782 Santiago de Compostela, Spain). Biomonitoring metal deposition in Galicia (NW Spain) with mosses: factors affecting bioconcentration. *Chemosphere*, 46(4) (2002), 535-542.

Three factors (canopy effect, lithology and seasonal variations) that may influence the concentrations of metals in terrestrial mosses were studied. The levels of 17 elements were determined in terrestrial mosses (*Scleropodium purum* (Hedw.) Limpr. and *Hypnum cupressiforme* Hedw.) collected from 75 sites in Galicia at two sampling times, in 1995 and 1997. In addition, monthly samples of *S. purum* were collected throughout a period of one year from four sites in the same area, for analysis of levels of eight elements. The first studied factor, collection of mosses from areas under tree cover, did not influence significantly the levels of the elements analysed. The second factor studied was the dominant lithology in the sampling area (granite, slate and schist). No significant differences were found between samples from sites where granites and slates dominated. Significant differences were found in the levels of Co, Cr, Ni and Mn in both species growing in granite and slate substrate areas when compared with those growing in schist areas. This was also found for Al and Fe in *S. purum* and for As in *H. cupressiforme*. The third factor investigated, using the results from the monthly survey, was the seasonal effect. No significant differences were found in the concentrations of all elements in *S. purum* throughout the year.

J. W. C. Wong, L. Xiang, X. Y. Gu, L. X. Zhou. (Department of Biology, Hong Kong Baptist University, Kowloon Tong, Hong Kong, PR China. Reaction Engineering Group, Department of Chemical Engineering, Tsinghua University, Beijing 100084,

PR China. Department of Microbiology, College of Life Science, Nanjing Agricultural University, Nanjing 210095, PR China. Department of Environmental Engineering, College of Resources and Environmental Sciences, Nanjing Agricultural University, Nanjing 210095, PR China). Bioleaching of heavy metals from anaerobically digested sewage sludge using FeS₂ as an energy source. *Chemosphere*, 55(1) (2004), 101-107.

The effect of using FeS₂ as an energy source, on the bioleaching of heavy metals (Zn, Cr, Cu, Pb and Ni) and nutrients (nitrogen and phosphorus) from anaerobically digested sludge using isolated indigenous iron-oxidizing bacteria was investigated in this paper. Addition of FeS₂ in the range of 0.5–4.0 g l⁻¹ accelerated the acidification of sludge and raised the oxidation–reduction potential of sludge medium with an inoculation of 15% (v/v) of active bacteria, thus resulting in an overall increase in metal dissolution efficiency. After 16 days of bioleaching at 28 °C and an initial pH of 3.0, up to 99% of Zn, 65% of Cr, 74% of Cu, 58% of Pb and 84% of Ni can be removed from the sludge. In contrast, only 94% of Zn, 12% of Cr, 21% of Cu, 32% of Pb and 38% of Ni were leached out in the control without inoculation of iron-oxidizing bacteria and the addition of FeS₂. Less than 15% of nitrogen and 6% of phosphorous were lost after 16 days of bioleaching when using FeS₂ as the energy source. Comparing to 39% and 45% loss respectively for these two nutrients when using FeSO₄ · 7H₂O as the energy source, FeS₂ appears to be a more suitable energy source for preserving nutrients in sludge while removing heavy metals from sludge.

Jeong-Hun Park, Yucheng Feng, Pingsheng Ji, Thomas C. Voice, Stephen A. Boyd. (Department of Civil and Environmental Engineering, Department of Crop and Soil Sciences, Michigan State University, East Lansing, Michigan 48824, Department of Agronomy and Soils, Auburn University, Auburn, Alabama 36849). Assessment of Bioavailability of Soil-Sorbed Atrazine. *Applied and Environmental Microbiology*, 69(6) (2003), 3288-3298.

Bioavailability of pesticides sorbed to soils is an important determinant of their environmental fate and impact. Mineralization of sorbed atrazine was studied in soil and clay slurries, and a desorption-biodegradation-mineralization (DBM) model was developed to quantitatively evaluate the bioavailability of sorbed atrazine. Three atrazine-degrading bacteria that utilized atrazine as a sole N source (*Pseudomonas* sp. strain ADP, *Agrobacterium radiobacter* strain J14a, and *Ralstonia* sp. strain M91-3) were used in the bioavailability assays. Assays involved establishing sorption equilibrium in sterile soil slurries, inoculating the system with organisms, and measuring the CO₂ production over time. Sorption and desorption isotherm analyses were performed to evaluate distribution coefficients and desorption parameters, which consisted of three desorption site fractions and desorption rate coefficients. Atrazine sorption isotherms were linear for mineral and organic soils but displayed some nonlinearity for K-saturated montmorillonite. The desorption profiles were well described by the three-site desorption model. In many instances, the mineralization of atrazine was accurately predicted by the DBM model, which accounts for the extents and rates of sorption/desorption processes and assumes biodegradation of liquid-phase, but not sorbed, atrazine. However, for the Houghton muck soil, which manifested the highest sorbed atrazine concentrations, enhanced mineralization rates, i.e., greater than those expected on the basis of aqueous-phase atrazine concentration, were observed. Even the assumption of instantaneous desorption could not account for the elevated rates. A plausible explanation for enhanced bioavailability is that bacteria access the localized regions where atrazine is sorbed and that the concentrations found support higher mineralization rates than predicted on the basis of aqueous-phase concentrations. Characteristics of high sorbed-phase concentration, chemotaxis, and attachment of cells to soil particles seem to contribute to the bioavailability of soil-sorbed atrazine.

Julio A. (Departamento Interuniversitario de Ecología, Edificio de Ciencias, Universidad de Alcalá, Alcalá de Henares, Madrid E-28871, Spain). Fluoride toxicity to aquatic organisms: a review. *Chemosphere*, 50(3), (2003), 251-264.

Published data on the toxicity of fluoride (F⁻) to algae, aquatic plants, invertebrates and fishes are reviewed. Aquatic organisms living in soft waters may be more adversely affected by fluoride pollution than those living in hard or seawaters because the bioavailability of fluoride ions is reduced with increasing water hardness. Fluoride can either inhibit or enhance the population growth of algae, depending upon fluoride concentration, exposure time and algal species. Aquatic plants seem to be effective in removing fluoride from contaminated water under laboratory and field conditions. In aquatic animals, fluoride tends to be accumulated in the exoskeleton of invertebrates and in the bone tissue of fishes. The toxic action of fluoride resides in the fact that fluoride ions act as enzymatic poisons, inhibiting enzyme activity and, ultimately, interrupting metabolic processes such as glycolysis and synthesis of proteins. Fluoride toxicity to aquatic invertebrates and fishes increases with increasing fluoride concentration, exposure time and water temperature, and decreases with increasing intraspecific body size and water content of calcium and chloride. Freshwater invertebrates and fishes, especially net-spinning caddisfly larvae and upstream-migrating adult salmons, appear to be more sensitive to fluoride toxicity than estuarine and marine animals. Because, in soft waters with low ionic content, a fluoride concentration as low as 0.5 mg F/l can adversely affect invertebrates and fishes, safe levels below this fluoride concentration are recommended in order to protect freshwater animals from fluoride pollution.

Kaijun Wang and Baoshan Xing. (Department of Plant and Soil Sciences, Stockbridge Hall, University of Massachusetts, P.O. Box 37245, Amherst, MA 01003-7245, USA). Adsorption and desorption of cadmium by goethite pretreated with phosphate. *Chemosphere*, 48(7) (2002), 665-670.

The adsorption of Cd by oxides or soils have been extensively studied, however, the desorption has received relatively limited attention, especially in the presence of phosphate. In this study, a batch equilibration method was used to investigate Cd sorption and desorption by goethite pretreated with phosphate. Phosphate not only enhanced Cd adsorption, but also accelerated the adsorption process. Compared with Cd adsorption by goethite alone, phosphate substantially moved the adsorption curves (edges) to lower pH range, indicative of enhancement of Cd sorption. The Cd adsorption by the pretreated goethite reached apparent equilibrium within 24 h at 20 °C, while such equilibrium was not observed after 4 weeks in the absence of phosphate. Cadmium was more readily released from phosphate-treated goethite. It is believed that phosphate blocked the pores on goethite surface, which lead to the fast adsorption kinetics and high extraction percentage. These results provided strong support for the diffusion of Cd into goethite particles.

Katrine Borgå, Bjørn Gulliksen, Geir Wing Gabrielsen, Janneche Utne Skaare. (Norwegian Polar Institute, N-9296 Tromsø, Norway. Norwegian College of Fishery Science, University of Tromsø, N-9037 Tromsø, Norway. Norwegian School of Veterinary Science, P.O. Box 8146, N-0033 Oslo, Norway. National Veterinary Institute, P.O. Box 8156 Dep., N-0033 Oslo, Norway). Size-related bioaccumulation and between-year variation of organochlorines in ice-associated amphipods from the Arctic Ocean. *Chemosphere*, 46(9-10) (2002), 1383-1392.

Organochlorines are found even in organisms living in remote areas such as the Arctic marginal ice zone. Organochlorine concentrations in ice-associated (sympagic) amphipods are related to their diet. Therefore, the size-dependent diet shift of *Gammarus wilkitzkii* may influence its organochlorine content. In this preliminary study, the organochlorine concentrations in two size classes (small ≤ 29 mm and large >29 mm) of *G. wilkitzkii* were

analysed. The concentrations of more lipophilic compounds [e.g. polychlorinated biphenyls (PCBs)] were lower in the small than the large size class, whereas concentrations of less lipophilic compounds [e.g. hexachlorocyclohexanes (HCHs)] did not differ by size class. Since contamination transport by the atmosphere, ocean currents and sea ice may vary, the organochlorine burden in sympagic organisms may also vary. There are no data available on temporal variation of organochlorine burden in Arctic sympagic fauna. Therefore, we compared organochlorine concentrations in sympagic amphipods (*G. wilkitzkii*, *Apherusa glacialis* and *Onisimus* spp.) between 1998 and 1999. Organochlorine concentrations in all amphipods were low in both years, with sum organochlorines from 50.7 to 621.9 ng g⁻¹ lipid weight. The concentrations of hexachlorobenzene (HCB), chlordanes, DDTs and PCBs were higher in 1999 than 1998, whereas the HCH concentrations were lower in 1999 than 1998. The organochlorine concentrations differed between the taxa in both years in a similar increasing manner from *A. glacialis* to both *G. wilkitzkii* and *Onisimus* spp. In studies of bioaccumulation relative to body size and temporal variation, a thorough interpretation requires samples from several size classes and years. Nevertheless, the present data provide new knowledge on contaminants in Arctic invertebrates where data are scarce.

Luciana Migliore, Salvatore, Cozzolino, Maurizio Fiori. (Dipartimento di Biologia, Università "Tor Vergata", Via della Ricerca Scientifica, Rome I-00133, Italy. Dipartimento di Biologia Vegetale, Università "Federico II", Via Foria 223, Napoli I-80139, Italy. Laboratorio di Medicina Veterinaria, Istituto Superiore di Sanità, Viale Regina Elena 229, Rome I-00161, Italy). Phytotoxicity to and uptake of enrofloxacin in crop plants. *Chemosphere*, 52(7) (2003), 1233-1244.

Phytotoxicity of enrofloxacin on crop plants *Cucumis sativus*, *Lactuca sativa*, *Phaseolus vulgaris* and *Raphanus sativus* was determined in a laboratory model: the effect of 50, 100 and 5000 µg l⁻¹ were evaluated after 30 days exposure by measuring post-germinative growth of primary root, hypocotyl, cotyledons and leaves. Concentrations between 50 and 5000 µg l⁻¹ induced both toxic effect and hormesis in plants, by significantly modifying both length of primary root, hypocotyl, cotyledons and the number/length of leaves. A toxic effect is induced by high concentration (5000 µg l⁻¹), while hormesis occurs at low concentrations (50 and 100 µg l⁻¹). A continuum between toxic effect and hormesis is found in the four plant species. Both toxic effect and hormesis can be related to an efficient plant drug uptake, in the order of µg g⁻¹. Plants are able to metabolize enrofloxacin into ciprofloxacin, as also happens in animals; *Cucumis*, *Lactuca* and *Phaseolus* biologically convert about one quarter of stored enrofloxacin. The ecological implication of enrofloxacin contamination in terrestrial environments is discussed.

Mahnaz Mazaheri Assadi, Maryam Mazaheri, Taher Nejad Satari, Ashrafulsadat Noohi, Manoochehr Shahamat, Morris Levin. Biosorption of Baftkar textile effluent. *Indian Journal of Experimental Biology*, 41 (2003), 900-904.

Decolourization of wastewater from a textile plant by a marine *Aspergillus niger* was studied. The fungus was previously isolated from Gorgan Bay in the Caspian Sea. The kinetics of decolourization was studied by varying energy sources. The best decolourization was achieved when sucrose was used as source of carbon and energy. NH ion was demonstrated to be the best nitrogen source. Color reduction was found to increase from 80-97% as inoculum concentration increased from 0.04-1.0g/L. A minimum inoculum of 0.2g/L is necessary to achieve decolourization. The optimal temperature for the growth of *A. niger* on Baftkar wastewater is found to be 30°C. 90-96% colour reduction is achieved in 19-20 hr of contact of mycelium cell with the wastewater. Colour reduction in a continuous column reactor of 70% was obtained using treated mycelium (NaOH, 90°C) after 1 hr.

Kumiko Miyoshi, Takehiko Nishio, Akio Yasuhara, Masatoshi Morita, Takayuki Shibamoto. (Institute of Applied Biochemistry, University of Tsukuba, 1-1-1

Tennodai, Tsukuba, Ibaraki 305-0006, Japan. Department of Chemistry, University of Tsububa, 1-1-1 Tennodai, Tsukuba, Ibaraki 305-0006, Japan. National Institute for Environmental Studies, 16-2 Onogawa, Tsukuba, Ibaraki 305-8506, Japan. Department of Environmental Toxicology, University of California, Meyer Hall, One Shields Avenue, Davis, CA 95616-8588, USA). Detoxification of hexachlorobenzene by dechlorination with potassium–sodium alloy. *Chemosphere*, 55(11) (2004), 1439-1446.

Dechlorination of hexachlorobenzene (HCB) was achieved by a liquid potassium–sodium (K–Na)-alloy. HCB in a cyclohexane/benzene solution (22 mmol/l, 4.67 g/l as chlorine) was dechlorinated by almost 100% after a 30-min reaction, indicating high reactivity of K–Na alloy and high proton donating power of cyclohexane. Decreasing orders of chlorobenzenes identified after a 15-min reaction, by amount were 1,2,3,4- > 1,2,3,5- > 1,2,4,5- for tetrachlorobenzenes, 1,2,4- > 1,2,3- > 1,3,5- for trichlorobenzenes, and 1,4- > 1,3- > 1,2- for dichlorobenzenes. It was hypothesized that once one chlorine atom in HCB was replaced with a proton, the adjacent chlorine atom to the proton tended to be replaced with another hydrogen atom. A total of 63 PCBs formed via the Wurtz–Fittig reaction were identified as by-products in the sample after a 15-min reaction. Among PCBs found, 2,3',4',5'-tetrachlorobiphenyl, which was a product from 1,2,4-trichlorobenzene formed via the Wurtz–Fittig reaction, was detected in relatively high concentration (48.9 nmol/ml). The sample obtained from a reaction mixture after 30 min contained only 14 PCBs in trace amounts, indicating that the PCBs formed were also further dechlorinated by K–Na alloy. Non-chlorinated compounds--such as methylbenzene, dimethylbenzene, dimer of tetrahydrofuran, and dicyclohexyl (dimer of cyclohexane)--were also identified in the samples. A method using K–Na alloy developed in the present study dechlorinated satisfactorily HCB at room temperature.

Mónica João de Barros Amorim, José Paulo Sousa, António J. A. Nogueira, Amadeu M. V. M. Soares. (Departamento de Biologia, Universidade de Aveiro, P-3810-193, Aveiro, Portugal. Instituto do Ambiente e Vida--Dpto. Zoologia da Universidade de Coimbra, P-3004-517, Coimbra, Portugal). Bioaccumulation and elimination of ¹⁴C-lindane by *Enchytraeus albidus* in artificial (OECD) and a natural soil. *Chemosphere*, 49(3) (2002), 323-329.

Bioaccumulation and elimination of ¹⁴C-lindane in *Enchytraeus albidus* was studied in artificial OECD soil and a silty loam from an agricultural field in Central West Portugal. Results showed that enchytraeids were able to bioaccumulate the chemical with a kinetic pattern similar to that of earthworms: fast uptake within a few days and a biphasic elimination pattern. A 10 day period to study uptake was sufficient, but a few more days were probably necessary for elimination. Bioaccumulation was influenced by soil type. The authors suggest that higher organic matter (OM) content and also the higher content on sand particles in the OECD soil may have led to a faster elimination: hydrophobic chemicals tend to adsorb to OM being in this way less bioavailable and therefore less bioaccumulated having bioaccumulation factor value around 6 while in natural soil is 10; the sand could act as abrasive particles (helpers) in the elimination process leading to an elimination of 90% of the chemical in two days while in natural soil 67% was eliminated in the same period of time.

Nico Boon, Eva M. Top, Willy Verstraete, Steven D. Siciliano. (Laboratory of Microbial Ecology and Technology (LabMET), Ghent University, B-9000 Ghent, Belgium). Bioaugmentation as a Tool To Protect the Structure and Function of an Activated-Sludge Microbial Community against a 3-Chloroaniline Shock Load. *Applied and Environmental Microbiology*, 69(3) (2003), 1511-1520.

Bioaugmentation of bioreactors focuses on the removal of xenobiotics, with little attention typically paid to the recovery of disrupted reactor functions such as ammonium-nitrogen removal. Chloroanilines are widely used in industry as a precursor to a variety of products and are occasionally released into wastewater streams. This work evaluated the effects on activated-sludge reactor functions of a 3-chloroaniline (3-CA) pulse and bioaugmentation by inoculation with the 3-CA-degrading strain *Comamonas testosteroni* I2 gfp. Changes in functions such as nitrification, carbon removal, and sludge compaction were studied in relation to the sludge community structure, in particular the nitrifying populations. Denaturing gradient gel electrophoresis (DGGE), real-time PCR, and fluorescent in situ hybridization (FISH) were used to characterize and enumerate the ammonia-oxidizing microbial community immediately after a 3-CA shock load. Two days after the 3-CA shock, ammonium accumulated, and the nitrification activity did not recover over a 12-day period in the nonbioaugmented reactors. In contrast, nitrification in the bioaugmented reactor started to recover on day 4. The DGGE patterns and the FISH and real-time PCR data showed that the ammonia-oxidizing microbial community of the bioaugmented reactor recovered in structure, activity, and abundance, while the number of ribosomes of the ammonia oxidizers in the nonbioaugmented reactor decreased drastically and the community composition changed and did not recover. The settleability of the activated sludge was negatively influenced by the 3-CA addition, with the sludge volume index increasing by a factor of 2.3. Two days after the 3-CA shock in the nonbioaugmented reactor, chemical oxygen demand (COD) removal efficiency decreased by 36% but recovered fully by day 4. In contrast, in the bioaugmented reactor, no decrease of the COD removal efficiency was observed. This study demonstrates that bioaugmentation of wastewater reactors to accelerate the degradation of toxic chlorinated organics such as 3-CA protected the nitrifying bacterial community, thereby allowing faster recovery from toxic shocks.

P. Adamo, M. Arienzo, M. Pugliese, V. Roca, P. Violante. (Dipartimento di Scienze del Suolo, della Pianta e dell'Ambiente, Università di Napoli Federico II, Via Università, 100-80055, Portici (NA), Italy. Dipartimento di Scienze Fisiche, Università di Napoli Federico II, Monte S. Angelo, Via Cintia, 80126, Napoli, Italy). Accumulation history of radionuclides in the lichen *Stereocaulon vesuvianum* from Mt. Vesuvius (south Italy). *Environmental Pollution*, 127(3) (2004), 455-461.

The fruticose lichen *Stereocaulon vesuvianum*, growing on the slopes of Mt. Vesuvius (south Italy), was used as a biomonitor of ^{134}Cs , ^{137}Cs , ^{103}Ru and ^{106}Ru derived from the April 26 1986 Chernobyl nuclear reactor accident. Samples were taken at five different quotes (370, 490, 580, 780 and 960 m a.s.l.) and four successive dates (October 1986, December 1986, October 1987 and May 1999). At the first sampling, the concentrations (as Bq kg^{-1} dry weight) ranged between 460 and 1020 for ^{134}Cs , 1330 and 2500 for ^{137}Cs , 90 and 200 for ^{103}Ru and 360 and 710 for ^{106}Ru , values generally lower in respect to those measured in soil and higher plants. Of the total ^{137}Cs measured only 14% was due to 1950s and 1960s nuclear weapons tests fallout. Highest average activities of all nuclides were observed at the quote of 960 m and significant correlation ($0.7 < r^2 < 0.9$) with altitude was found for ^{106}Ru and ^{137}Cs values. The time span between the first two samplings (38 days) was enough to highlight only the decay of ^{103}Ru ($T_{1/2}=40$ days). At sampling performed 1 year later (October 1987) only ^{134}Cs , ^{137}Cs , and ^{106}Ru were observed. Thirteen years after the Chernobyl incident (May 1999), although a decrease was obvious with time, the ^{137}Cs content in lichen thallus was still appreciable ($270\text{--}570 \text{ Bq kg}^{-1} \text{ dw}$). Experimental results from the first three samplings yield effective residence time of 0.98 ± 0.07 , 5.9 ± 0.3 and 1.6 ± 0.1 years for ^{106}Ru , ^{137}Cs and ^{134}Cs , respectively. The last sampling of May 1999 allowed to achieve a more precise determination of lifetime of ^{137}Cs (6.1 ± 0.4 years).

Pairat Kaewsarn. (Department of Chemical Engineering, Ubon Ratchathani University, Warinchamrab, Ubon Ratchathani 34190, Thailand). Biosorption of copper(II) from aqueous solutions by pre-treated biomass of marine algae *Padina* sp. *Chemosphere*, 47(10) (2002), 1081-1085.

Biosorption of heavy metals can be an effective process for the removal and recovery of heavy metal ions from aqueous solutions. The biomass of marine algae has been reported to have high uptake capacities for a number of heavy metal ions. In this paper, the adsorption properties of a pre-treated biomass of marine algae *Padina* sp. for copper(II) were investigated. Equilibrium isotherms and kinetics were obtained from batch adsorption experiments. The biosorption capacities were solution pH dependent and the maximum capacity obtained was 0.80 mmol/g at a solution pH of about 5. The biosorption kinetics was found to be fast, with 90% of adsorption within 15 min and equilibrium reached at 30 min. The effects of light metal ions on copper(II) uptake were studied and the presence of light metal ions did not affect copper(II) uptake significantly. Fixed-bed breakthrough curves for copper(II) removal were also obtained. This study demonstrated that the pre-treated biomass of *Padina* sp. could be used as an effective biosorbent for the treatment of copper(II) containing wastewater streams.

Patricia A. Terry, Wendy Stone. (Department of Natural and Applied Sciences, University of Wisconsin--Green Bay, ES 317, 2420 Nicolet Drive, Green Bay, WI 54311, USA). Biosorption of cadmium and copper contaminated water by *Scenedesmus abundans*. *Chemosphere*, 47(3) (2002), 249-255.

Experiments were conducted comparing the individual removals of cadmium and copper from water via biosorption using *Scenedesmus abundans*, a common green algae, to removal in a multi-component system to determine competitive effects, if any, between the metals. The goal was to characterize the biological treatment of water contaminated with heavy metals using live aquatic species. In addition, experiments were performed to measure cell viability as a function of metal concentration and also to compare metal removal using living species to that using nonliving ones. It was shown that, while both living and nonliving *S. abundans* removed cadmium and copper from water, living algae significantly outperformed nonliving algae. Further, in characterizing biosorption by three concentrations of live *S. abundans*, capacity curves were created comparing the metal biosorbed per mass algae to the initial metal concentration in solution. The algae concentration was not a factor in the biosorption of either metal individually, such that the capacity of the algae for the metal increased with decreasing algae concentration. At the lowest algae concentration considered, competitive effects were observed at copper and cadmium concentrations above 4 mg/l each. At the highest algae concentration considered, no competitive effects were observed in the range of cadmium and copper concentrations studied (1–7 mg/l). It was concluded that biological treatment of heavy metal contaminated water is possible and that at adequately high algae concentrations, multi-component metal systems can be remediated to the same level as individual metals.

Peter F. Landrum, Guilherme R. Lotufo, Duane C. Gossiaux, Michelle L. Gedeon, Jong-Hyeon Lee. (Great Lakes Environmental Research Laboratory, NOAA 2205 Commonwealth Blvd., Ann Arbor, MI, 48105, USA. US Army Engineer Research and Development Center, 3909 Halls Ferry Rd., Vicksburg, MS 39180, USA. Cooperative Institute for Limnology and Ecosystem Research, University of Michigan, Ann Arbor, MI 48109, USA. Department of Oceanography, Seoul National University, Seoul 151-742, South Korea). Bioaccumulation and critical body residue of PAHs in the amphipod, *Diporeia* spp.: additional evidence to support toxicity additivity for PAH mixtures. *Chemosphere*, 51(6) (2003), 481-489.

Polycyclic aromatic hydrocarbons (PAHs) are considered to act additively when exposed as congener mixtures. Additive internal concentrations at the site of toxic action is the basis for recent efforts to establish a sum PAH guideline for sediment-associated PAH toxicity. This study determined the toxicity of several PAH congeners on a body residue basis in *Diporeia* spp. These values were compared to the previously established LR₅₀ value for a PAH mixture based on the molar sum of PAH congeners and demonstrated similar LR₅₀ values for individual PAH. These results support the contention that the PAH act at the same molar concentration whether present as individual compounds or in mixture. Aqueous exposures were conducted for 28 d, and the water was exchanged daily to maintain the exposure concentration. The concentration in the exposures declined by an average of 22% between water exchanges across all compounds, and ranged from 11% to 32%. The toxicokinetics were determined using both time-weighted-average (TWA) and time-variable water concentrations and were not statistically different between the two source functions. Toxicity was determined for both mortality and immobility (failure to swim on prodding) and on both a TWA water concentration and a body residue basis. The LC₅₀ values ranged from 1757 µg l⁻¹ for naphthalene after 10 d exposure to 79.1 µg l⁻¹ for pyrene after 28 d exposure, and the EC₅₀ ranged from 1587 µg l⁻¹ for naphthalene after 10 d exposure to 38.2 µg l⁻¹ for pyrene after 28 d exposure. The LR₅₀ values for all congeners at all lengths of exposure were essentially constant and averaged 7.5±2.6 µmol g⁻¹, while the ER₅₀ for immobility averaged 2.6±0.6 µmol g⁻¹. The bioconcentration factor declined with increasing exposure concentration and was driven primarily by a lower uptake rate with increasing dose, while the elimination remained essentially constant for each compound.

PK Gill, DS Arora, M Chander. (Microbial Technology Laboratory, Department of Microbiology, Guru Nanak Dev University, Amritsar, Punjab, 143 005 India). Biodecolourization of azo and triphenylmethane dyes by *Dichomitus squalens* and *Phlebia* spp. *Journal of Industrial Microbiology and Biotechnology*, 28(4) (2002), 201 – 203.

Nine white-rot fungal strains were screened for biodecolourization of brilliant green, cresol red, crystal violet, congo red and orange II. *Dichomitus squalens*, *Phlebia fascicularia* and *P. floridensis* decolourized all of the dyes on solid agar medium and possessed better decolourization ability than *Phanerochaete chrysosporium* when tested in nitrogen-limited broth medium.

Seiji Katano, Yuka Matsuo, Ken'ichi Hanaoka. (Katano Bussan Co. Ltd., Mukunomachi 3-13-18, Shimonoseki 751-0816, Japan. Department of Food Science and Technology, National Fisheries University, Nagata-honmachi 2-7-1, Shimonoseki 759-6595, Japan). Arsenic compounds accumulated in pearl oyster *Pinctada fucata*. *Chemosphere*, 53(3) (2003), 245-251.

We investigated the water-soluble arsenic compounds present in the soft tissues of both the pearl-free and the pearl-containing pearl oysters. After dividing the soft tissue into five parts, i.e., adductor muscle, foot, mantle, viscera and gill, each part was analyzed by high-performance liquid chromatography–inductively coupled plasma mass spectrometry for the arsenic compounds accumulated in it. Arsenic concentration of each tissue part ranged from 22.1 to 45.7 µg g⁻¹ of dry tissue in the pearl-free pearl oyster and from 27.4 to 50.4 µg g⁻¹ of dry tissue in the pearl-containing pearl oyster. On the grounds of the present evidence the major water-soluble arsenic compound accumulated in each part was identified as arsenobetaine without exception in both types of pearl oysters (94.3–99.7% in the pearl-free pearl oyster and 87.2–99.7% in the pearl-containing pearl oyster). Trace or small amounts of arsenic compounds including tetramethylarsonium ion and arsenocholine were also detected in some parts. The levels of these minor arsenicals were a little higher in pearl-free pearl oyster than in the pearl-containing pearl oyster. This study confirms the

hygienic safety of the soft tissues of both the pearl-free and the pearl-containing pearl oysters, as food.

Sinha S, Saxena R, and Singh S. (Ecotoxicology and Bioremediation, Environmental Sciences Division, National Botanical Research Institute, Lucknow, India). Comparative studies on accumulation of Cr from metal solution and tannery effluent under repeated metal exposure by aquatic plants: Its toxic effects. Environmental Monitoring and Assessment, 80(1) (2002), 17-31.

The present study demonstrates comparison of Cr accumulating potential by the plants of *Najas indica* Cham. (submerged), *Vallisneria spiralis* L. (rooted submerged) and *Alternanthera sessilis* R. Br. (rooted emergent) under repeated metal exposure and its effect on chlorophyll and protein concentrations. These plants were treated with different concentrations of Cr under repeated exposure in controlled laboratory conditions to assess the maximum metal accumulation potential. The plants of *V. spiralis* accumulated significantly high amount of Cr under laboratory conditions in comparison to *N. indica* and *A. sessilis*. These plants have shown a decrease in chlorophyll and protein concentrations with increase in Cr concentrations. In view of high accumulation of Cr in *V. spiralis*, the plants were treated with different concentrations of tannery effluent collected from Common Effluent Treatment Plant, Unnao (UP). The plants of *V. spiralis* treated with 100% tannery waste water showed the maximum accumulation ($57.5 \mu\text{g g}^{-1} \text{ dw}$) of Cr in the roots a days of exposure. The plants were found effective in removing Cr from solution and tannery effluent.

Suhendrayatna, Akira Ohki, Tsunenori Nakajima, Shigeru Maeda. (Department of Applied Chemistry and Chemical Engineering, Faculty of Engineering, Kagoshima University, 1-21-40 Korimoto, Kagoshima 890-0065, Japan. Department of Bioengineering, Faculty of Engineering, Kagoshima University, 1-21-40 Korimoto, Kagoshima 890-0065, Japan. Kagoshima National College of Technology, 1460-1 Shinkou, Hayato-cho, Kagoshima 899-5193, Japan). Studies on the accumulation and transformation of arsenic in freshwater organisms II. Accumulation and transformation of arsenic compounds by *Tilapia mossambica*. Chemosphere, 46(2) (2002), 325-331.

Bioaccumulation and biotransformation of arsenic (As) compounds in freshwater *Tilapia mossambica* was investigated. The direct accumulation of As by *T. mossambica* was proportional to the concentration of arsenicals in water. Small amounts of accumulated As were transformed to methylated As, including trimethylarsenic (TMA) species. Accumulation and transformation of As(III) by *T. mossambica* via freshwater food chain results in the transformation of As(III) to As(V) with little biomethylation of accumulated As. Approximately 90% of accumulated As was depurated to water.

Sven Burreau, Yngve Zebühr, Dag Broman, Rasha Ishaq. (Department of Zoology, Stockholm University, Stockholm SE-106 91, Sweden. Institute of Applied Environmental Research, Stockholm University, Stockholm SE-106 91, Sweden). Biomagnification of polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers (PBDEs) studied in pike (*Esox lucius*), perch (*Perca fluviatilis*) and roach (*Rutilus rutilus*) from the Baltic Sea. Chemosphere, 55(7) (2004), 1043-1052.

Pike, perch and roach from rural waters of the Baltic Sea were investigated for possible biomagnification of polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers (PBDEs). For this we used data on $\delta^{15}\text{N}$, weight and sex of the fish. We were able to separate body size effects from trophic position effects on biomagnification. Both these parameters lead to biomagnification of PCBs and PBDEs. All investigated PCBs (tri- to deca-

CBs) biomagnify and the biomagnification potential is positively correlated with hydrophobicity up to logKow 8.18. Tri- to hepta-BDEs also biomagnify but showed a maximum biomagnification for the penta-BDEs (logKow 6.46–6.97). The biomagnification of hexa- to hepta-PBDEs was negatively correlated with degree of bromination, likely due to large molecular size or high molecular weight (644–959 Da). Octa-, nona- and deca-BDEs did not biomagnify but were found in two (octa-BDE) and three (nona- and deca-BDEs) of the species, respectively. Increased size of pike is correlated with increased lipid weight based PCB and PBDE concentrations in males but not in females and mean PCB and PBDE concentrations in males are generally higher than in females. For the least hydrophobic PCBs, no sex difference is observed, probably as a consequence of faster clearance of these substances over the gills, making the spawning clearance of PCBs and PBDEs of lesser relative importance.

T. Manios, E. I. Stentiford, P. Millner. (School of Agricultural Technology, Technological and Educational Institute of Crete, Technical University of Crete, Heraklion 71500, Crete, Greece. School of Civil Engineering, Leeds University, LS2 9JT, Leeds, UK. School of Biochemistry and Molecular Biology, Leeds University, LS2 9JT, Leeds, UK). Removal of heavy metals from a metaliferous water solution by *Typha latifolia* plants and sewage sludge compost. *Chemosphere*, 53(5) (2003).

487-494. *Typha latifolia* plants, commonly known as cattails, were grown in a mixture of mature sewage sludge compost, commercial compost and perlite (2:1:1 by volume). Four Groups (A, B, C and D) were irrigated (once every two weeks) with a solution containing different concentrations of Cu, Ni, and Zn, where in the fifth (group M) tap water was used. At the end of the 10 weeks experimental period substrate and plants were dried, weighed and analysed for heavy metals. The amounts of all three metals removed from the irrigation solution, were substantial. In the roots and leaves/stems of *T. latifolia* the mean concentration of Zn reached values of 391.7 and 60.8 mg/kg of dry weight (d.w.), respectively. In the substrate of Group D all three metals recorded their highest mean concentrations of 1156.7 mg/kg d.w. for Cu, 296.7 mg/kg d.w. for Ni and 1231.7 mg/kg d.w. for Zn. Linear correlation analyses suggested that there was a linear relationship between the concentration of metals in the solutions and the concentration of metals in the substrates at the end of the experiment. The percentage removal of the metals in the substrate was large, reaching 100% for Cu and Zn in some groups and almost 96% for Ni in group D. The total amount of metals removed by the plants was considerably smaller than that of the substrate, due mainly to the small biomass development. A single factor ANOVA test (5% level) indicated that the build up in the concentration of metals in the roots and the leaves/stems was due to the use of metaliferous water solution and not from the metals pre-existing in the substrate. The contribution of the plants (both roots and leaves/stems) in the removing ability of the system was less than 1%.

T. Srinath, T. Verma, P. W. Ramteke, S. K. Garg. (Environmental Microbiology Section, Industrial Toxicology Research Centre, P.O. Box 80, M.G. Road, Lucknow 226 001, India. Department of Microbiology, Dr. R.M.L. Avadh University, Faizabad 224 001, India). Chromium (VI) biosorption and bioaccumulation by chromate resistant bacteria. *Chemosphere*, 48(4) (2002), 427-435.

In this study, strains that are capable of bioaccumulating Cr(VI) were isolated from treated tannery effluent of a common effluent treatment plant. The Cr(VI) concentration in this treated effluent was 0.96 mg/l, much above the statutory limit of 0.1 mg/l for discharge of industrial effluents into inland surface waters in India. In addition to the bioaccumulation, biosorption capabilities of living and dead cells were analysed. Two strains, identified as *Bacillus circulans* and *Bacillus megaterium* were able to bioaccumulate 34.5 and 32.0 mg

Cr/g dry weight, respectively and brought the residual concentration of Cr(VI) to the permissible limit in 24 h when the initial concentration was 50 mg Cr(VI)/l. Our experimental design accounts for initial as well as final residual concentration of heavy metal while selecting heavy metal accumulating strains during batch studies. Biosorption of Cr(VI) was shown by *B. megaterium* and another strain, *B. coagulans*. Living and dead cells of *B. coagulans* biosorbed 23.8 and 39.9 mg Cr/g dry weight, respectively, whereas, 15.7 and 30.7 mg Cr/g dry weight was biosorbed by living and dead cells of *B. megaterium*, respectively. Biosorption by the dead cells was higher than the living cells. This was due to prior pH conditioning (pH 2.5 with deionized water acidified with H₂SO₄) of the dead cells.

Tatsuya Ueki, Yasuhisa Sakamoto, Nobuo Yamaguchi, and Hitoshi Michibata. (Marine Biological Laboratory, Graduate School of Science, Hiroshima University, Hiroshima 722-0073, Japan). Bioaccumulation of Copper Ions by *Escherichia coli* Expressing Vanabin Genes from the Vanadium-Rich Ascidian *Ascidia sydneiensis samea*. Applied and Environmental Microbiology, 69(12) (2003), 7035-7043.

The genes encoding two vanadium-binding proteins, vanabin1 and vanabin2, from a vanadium-rich ascidian, *Ascidia sydneiensis samea*, were recently identified and cloned (T. Ueki, T. Adachi, S. Kawano, M. Aoshima, N. Yamaguchi, K. Kanamori, and H. Michibata, *Biochim. Biophys. Acta* 1626:43-50, 2003). The vanabins were found to bind vanadium(IV), and an excess of copper(II) ions inhibited the binding of vanadium(IV) to the vanabins in vitro. In this study, we constructed *Escherichia coli* strains that expressed vanabin1 or vanabin2 fused to maltose-binding protein (MBP) in the periplasmic space. We found that both strains accumulated about twenty times more copper(II) ions than the control BL21 strain, while no significant accumulation of vanadium was observed. The strains expressing either MBP-vanabin1 or MBP-vanabin2 absorbed approximately 70% of the copper ions in the medium to which 10 µM copper (II) ions were initially added. The MBP-vanabin1 and MBP-vanabin2 protein expressed in the periplasm bound to copper ions at a copper:protein molar ratio of 8:1 and 5:1, respectively, but MBP did not bind to copper ions. These data showed that the metal-binding proteins vanabin1 and vanabin2 bound copper ions directly and enhanced the bioaccumulation of copper ions by *E. coli*.

Tetsuo Ando, Megumi Yamamoto, Takashi Tomiyasu, Jun Hashimoto, Tomoyuki Miura, Atsuhiko Nakano, Suminori Akiba. (Department of Public Health, Faculty of Medicine, Kagoshima University, 8-35-1 Sakuragaoka, Kagoshima 890-8520, Japan. Department of Basic Medical Sciences, National Institute for Minamata Disease, 4058-18 Hama, Minamata, Kumamoto 867-0008, Japan. Department of Earth and Environmental Science, Faculty of Science, Kagoshima University, 1-21-35 Kohrimoto, Kagoshima 890-0065, Japan. Marine Ecosystems Research Department, Japan Marine Science and Technology Center, 2-15 Natsushima, Yokosuka, Kanagawa 237-0061, Japan. Faculty of Agriculture, Miyazaki University, 1-1 Gakuen-Kibanadai-Nishi, Miyazaki 889-2192, Japan). Bioaccumulation of mercury in a vestimentiferan worm living in Kagoshima Bay, Japan. Chemosphere, 49(5) (2002), 477-484.

The present study reports on the mercury concentrations of the vestimentiferan worm, *Lamellibrachia satsuma*, (Annelida: Pogonophora) found near hydrothermal vents at a depth of 80–100 m in the northern parts of Kagoshima Bay. The vestimentiferan worms had total mercury concentrations of 238 ng/g in the anterior muscle of the body and 164 ng/g in the posterior trophosome. Methylmercury constituted only 7.6% of total mercury detected anteriorly and 16.3% posteriorly. The mean total mercury concentration in filtrated ambient seawater of the worm habitat was 1.1 ng/l. The worm should accumulate mercury in seawater by a one-step into the anterior and posterior parts as 2.2×10^5 and 1.5×10^5 times those of the filtered ambient seawater, respectively. The bioaccumulation factor of mercury

by the worms with only their respiration would be actually larger than that by other marine animals through food webs. The high bioaccumulation factor of mercury in the worms suggest the following two possibilities: (i) the biological half-life of organomercury in the worm could be exceptionally long; or (ii) the lifetime of vestimentiferan worms examined in the present study could be extremely long. Various metals in one specimen of the worm were analyzed by using ICP-MS, and then gold as well as silver were detected in the worm. Gold was detected for the first time from marine animals.

Tiina Petänen, Martin Romantschuk. (Department of Biosciences, Division of General Microbiology, University of Helsinki, P.O. Box 56, FIN-00014, Finland. Department of Ecological and Environmental Sciences, University of Helsinki, Niemenkatu 73, FIN-15140, Lahti, Finland). Toxicity and bioavailability to bacteria of particle-associated arsenite and mercury. *Chemosphere*, 50(3) (2003), 409-413.

The overall toxicity of soil, and the bioavailability and arsenite from soil were measured with the constructed constitutively luminescent strain *Pseudomonas fluorescens* OS8 (pNEP01) and with earlier published biosensor strains *P. fluorescens* OS8 (pTPT11) for mercury and *P. fluorescens* OS8 (pTPT31) for arsenite, respectively. Both spiked and authentic samples were studied. By combining bacterial assays enabled partial analysis of reasons for toxicity of environmental samples, some of which were highly toxic despite containing little or no heavy metals. The spiked soils were not toxic overall but the method of measuring concentration from water-extractable fraction or from soil-water slurry affected the results significantly. Mercury that was bound to clay even after water extraction was nevertheless found to be bioavailable to a high degree to the biosensor bacteria. Since induction of the luminescence genes takes place intracellularly the bacteria may be able to apparently release mercury when in direct contact with clay particle. This type of biomobilisation was not observed with arsenite spiked soils. The same phenomenon was detected in one of the environmental samples.

Xin Hu, Zhuhong Ding, Yijun Chen, Xiaorong Wang, Lemei Dai. (State Key Laboratory of Pollution Control and Resource Reuse, School of Environment, Nanjing University, Nanjing 210093, China. Center of Material Analysis of Nanjing University, Nanjing 210093, China). Bioaccumulation of lanthanum and cerium and their effects on the growth of wheat (*Triticum aestivum* L.) seedlings. *Chemosphere*, 48(6) (2002), 621-629.

Through short-term exposure (7-d exposure), long-term exposure (16-d exposure) and exposure-recovery (7-d exposure+9-d recovery), the bioaccumulation and distribution of La and Ce and their effects on growth of wheat seedlings were studied. Addition of La (0.5–25 mg/l) and Ce (0.5–25 mg/l) to the culture medium individually and in combination inhibited primary root elongation, reduced the dry weight of roots and shoots and the content of mineral elements (Ca, Mg, K, Cu, Zn). The damage increased with an increase in the concentrations of La and Ce in culture medium. Relative damage ratio increased with an increase in concentrations of La and Ce in the culture medium and with exposure time. Comparing exposure-recovery groups with long-term exposure groups, primary root lengths, dry weight of roots and shoots and the content of five mineral elements were higher. The accumulation of La and Ce in the seedlings was positively correlated with the concentrations of La and Ce in the culture medium and with exposure time. Bioaccumulation factors of La and Ce in roots were much higher than those of shoots. The uptake rates of La and Ce by the plants were much higher than the translocation rates from roots to shoots. The accumulation and distribution of La and Ce in the seedlings in exposure-recovery groups showed that there was very little excretion through metabolism during the recovery

period, but redistribution occurred throughout the whole plant. No apparent selective uptake was found between La and Ce by the plants when they were applied in combination.

Youn-Seok Kang, Masumi Yamamuro, Shigeki Masunaga, Junko Nakanishi. (CREST, Japan Science and Technology Corporation, 4-1-8 Honcho, Kawaguchi, Saitama 332-0012, Japan. Marine Geology Department, Geological Survey of Japan, 1-1-3 Higashi, Tsukuba, Ibaraki 305-8567, Japan. Institute of Environmental Science and Technology, Yokohama National University, 79-7 Tokiwadai, Hodogaya, Yokohama 240-8501, Japan). Specific biomagnification of polychlorinated dibenzo-p-dioxins and dibenzofurans in tufted ducks (*Aythya fuligula*), common cormorants (*Phalacrocorax carbo*) and their prey from Lake Shinji, Japan. *Chemosphere*, 46(9-10) (2002), 1373-1382.

Polychlorinated dibenzo-p-dioxins and dibenzofurans (PCDDs/DFs) were detected in waterfowl such as common cormorants, tufted ducks, and their prey, namely fish and bivalves from Lake Shinji, Japan. The concentration of total PCDDs/DFs-TEQ was found to be higher in the muscle tissues of common cormorants than in those of tufted ducks. The results of hierarchical cluster analysis implied that the residue distribution pattern of PCDD/DF homologues was considerably different between these two species. Furthermore, biomagnification factors (BMFs) were estimated from bivalves as prey to tufted duck muscles as target organs. Despite the highest concentrations of 1,3,6,8- and 1,3,7,9-TeCDD in tufted ducks and their prey, however, the BMFs of these isomers were calculated to be lower than those of the toxic 2,3,7,8-substituted PCDDs/DFs. On the other hand, log BMF of toxic 2,3,7,8-substituted PCDDs/DFs were significantly higher for lower chlorinated isomers than those of the higher chlorinated isomers. The biota-sediment accumulation factors (BSAFs) of PCDDs/DFs were also estimated using shijimi clam and fish samples against sediment from Lake Shinji. The average BSAFs were estimated and ranged from 4.0×10^{-3} to 2.2×10^{-1} and 2.0×10^{-4} to 2.0×10^{-1} for bivalve and fish samples, respectively. Based on calculated BMFs and BSAFs, the total PCDD/DF-TEQ levels in the tufted duck were estimated to have been lowest (2.0 pg TEQ/g dry weight basis) in 1947 and highest (9.8 pg TEQ/g) in 1971.

Z. H. Ye, A. J. M. Baker, M. H. Wong, A. J. Willis. (Department of Biology, Institute for Natural Resource and Environmental Management, Hong Kong Baptist University, Kowloon Tong, Hong Kong SAR, PR China. School of Botany, The University of Melbourne, VIC 3010, Melbourne, Australia. Department of Animal and Plant Sciences, University of Sheffield, Sheffield S10 2TN, S. Yorks, UK). Copper tolerance, uptake and accumulation by *Phragmites australis*. *Chemosphere*, 50(6) (2003), 795-800.

Copper (Cu) uptake and accumulation of five populations of *Phragmites australis* growing in two mine sites (Parys Mountain, Wales, UK and Plombières, Belgium) contaminated with Cu and three 'clean' sites (Felixstowe, UK; Wisbech, UK and Mai Po, Hong Kong) were studied under field and glasshouse conditions. Cu tolerances in these populations of seedlings raised from seeds collected from the above five sites were also studied under glasshouse conditions. Although concentrations of Cu in the plant tissues (leaves, stems, rhizomes and roots) of *P. australis* and the associated soils from the Cu-contaminated sites were significantly higher than those of the plant tissues and the soils collected in the clean sites, small differences were found between the two Cu-contaminated populations and the three clean populations when seedlings were grown in 0.1 and 0.5 $\mu\text{g ml}^{-1}$ Cu treatment solutions. In general, different populations of seedlings showed similar growth responses, metal uptake and indices of Cu tolerance when cultured in the same Cu treatment solution for 3 weeks. There was insufficient evidence to support the hypothesis that the Cu-contaminated populations have evolved to Cu-tolerant ecotypes.

Zümriye Aksu, Gönül Dönmez. (Department of Chemical Engineering, Hacettepe University, 06532, Beytepe, Ankara, Turkey. Department of Biology, Ankara University, 06100, Ankara, Turkey). A comparative study on the biosorption characteristics of some yeasts for Remazol Blue reactive dye. Chemosphere, 50(8) (2003), 1075-1083.

Biosorption capacities and rates of different kinds of dried yeasts (*Saccharomyces cerevisiae*, *Schizosaccharomyces pombe*, *Kluyveromyces marxianus*, *Candida* sp., *C. tropicalis*, *C. lipolytica*, *C. utilis*, *C. quilliermendii* and *C. membranaefaciens*) for Remazol Blue reactive dye from aqueous solutions were compared under laboratory conditions as a function of initial pH and initial dye concentration. Optimum initial biosorption pH was determined as 2 for all the yeasts. All the yeast species showed comparable and very high dye sorption at 100 mg/l initial dye concentration. The equilibrium sorption capacity of the biomass increased with increasing initial dye concentration up to 400 mg/l for *Candida* sp. *C. lipolytica* and *C. tropicalis*; up to 300 mg/l for *C. quilliermendii* and *C. utilis* and up to 200 mg/l for *S. cerevisiae*, *S. pombe*, *K. marxianus* and *C. membranaefaciens* while the adsorption yield of dye showed the opposite trend for all the yeasts. Among the nine yeast species, *C. lipolytica* exhibited the highest dye uptake capacity ($Q^0=250$ mg/g). Both the Freundlich and Langmuir adsorption models were found suitable for describing the biosorption of the dye by all the *Candida* yeasts (except *C. membranaefaciens*). The results indicated that the dye uptake process followed the pseudo-second-order kinetics for each dye-yeast system.

Biodegradation

A. Bahrami, S.A. Shojaosadati, G. Mohebal. (Biotechnology Group, Chemical Engineering Department, Tarbiat Modarres University, Tehran, Iran). Biodegradation of dibenzothiophene by thermophilic bacteria. Biotechnology Letters, 23(11) (2001), 899-901.

Anaerobic microbial biodegradation of dibenzothiophene (DBT) was studied using thermophilic bacteria obtained from crude oil. A mixed culture was obtained that degraded 98% of DBT at 0.5 mg ml⁻¹ at 65 °C over 15 days both in the presence and in the absence of Methyl Viologen.

Adolf Eisentraeger, Martin Schmidt, Hubertus Murrenhoff, Wolfgang Dott, Stefan Hahn. (Institute of Hygiene and Environmental Medicine, RWTH Aachen, Pauwelsstrasse 30, D-52057 Aachen, Germany. Institute of Fluidpower Transmission and Control, Steinbachstrasse 53, D-52056 Aachen, Germany). Biodegradability testing of synthetic ester lubricants--effects of additives and usage. Chemosphere, 48(1) (2002), 89-96.

The optimised biodegradability test system " O_2/CO_2 Headspace Test with GC-TCD" is used for the assessment of synthetic ester lubricants. The effects of both additives and usage on biodegradability are examined and discussed. Ester based cutting fluids and hydraulic fluids with and without additives are used under defined conditions at machine tools and hydraulic and plain bearing test benches. The lubricants are characterised additionally with respect to kinematic viscosity, acidity and elemental composition. Furthermore, a formulated mineral oil is characterised before and after usage at an hydraulic test bench. The results clearly show that the mineral oil is far less biodegradable than the ester oils and that their biodegradability is not affected by usage. Biodegradability of the ester oils is mainly depending on the characteristics of the base fluids and not affected by the additives. Antioxidants are influencing stability respectively biodegradability indirectly, since they prevent oxopolymerisation effects. Other effects of usage on biodegradation are not

detected. In this context, the antioxidants ensure ready biodegradability and have a positive effect on the environmental fate of synthetic ester lubricants.

Agnes Pierwola, Tomasz Krupinski, Peter Zalupski, Michael Chiarelli, Domenic Castignetti. (Departments of Biology,¹ Chemistry, Loyola University of Chicago, Chicago, Illinois 60626). Degradation Pathway and Generation of Monohydroxamic Acids from the Trihydroxamate Siderophore Deferrioxamine B. Applied and Environmental Microbiology, 70(2) (2004), 831-836.

Siderophores are avid ferric ion-chelating molecules that sequester the metal for microbes. Microbes elicit siderophores in numerous and different environments, but the means by which these molecules reenter the carbon and nitrogen cycles is poorly understood. The metabolism of the trihydroxamic acid siderophore deferrioxamine B by a *Mesorhizobium loti* isolated from soil was investigated. Specifically, the pathway by which the compound is cleaved into its constituent monohydroxamates was examined. High-performance liquid chromatography and mass-spectroscopy analyses demonstrated that *M. loti* enzyme preparations degraded deferrioxamine B, yielding a mass-to-charge (*m/z*) 361 dihydroxamic acid intermediate and an *m/z* 219 monohydroxamate. The dihydroxamic acid was further degraded to yield a second molecule of the *m/z* 219 monohydroxamate as well as an *m/z* 161 monohydroxamate. These studies indicate that the dissimilation of deferrioxamine B by *M. loti* proceeds by a specific, achiral degradation and likely represents the reversal by which hydroxamate siderophores are thought to be synthesized.

Akio Tsuchii, Yutaka Tokiwa. (Research Institute of Biological Resources, National Institute of Advanced Industrial Science and Technology, Higashi 1-1-3, Tsukuba, Ibaragi 305, Japan). Microbial degradation of tyre rubber particles. Biotechnology Letters, 23(12) (2001), 963-969.

Degradation of rubber particles from tyre treads, having diameters from 0.8 to 2.3 mm, was achieved using *Nocardia* sp. 835A-Rc, a mutant strain with strong rubber-degrading ability. The entire surface of the particles was uniformly attacked by the organism either without stirring of the culture medium or at a very low stirring rate of 40 rpm. At a higher rate of stirring, however, a small number of large microbial colonies were formed on the rubber surface and separate deep semi-spherical cavities were observed after the removal of microbial cells by washing. The number of microbial colonies decreased with increasing stirring rate but each one of the colonies became larger at the same time. As the result of these two counteracting effects of stirring on microbial activity, the weight loss of the particles increased when the stirring rate was raised from 0 to 40 rpm but decreased when the rate was increased from 40 to 70 or 150 rpm. At the stirring rate of 40 rpm, the weight losses of the particles with mean diameters of about 0.8, 1.1 and 2.3 mm were 57, 50 and 36%, respectively, after 8 weeks. The rate of microbial degradation increased again when the stirring was raised from 150 to 300 rpm.

Alfredo A. Marchetti, Mark G. Knize, Marina L. Chiarappa-Zucca, Ronald J. Pletcher and David W. Layton. (Environmental Science Division, Lawrence Livermore National Laboratory, 7000 East Avenue, L-396, Livermore, CA 94550, USA). Biodegradation of potential diesel oxygenate additives: dibutyl maleate (DBM), and tripropylene glycol methyl ether (TGME). Chemosphere, 52(5) (2003), 861-868.

The addition of oxygen-bearing compounds to diesel fuel considerably reduces particulate emissions. TGME and DBM have been identified as possible diesel additives based on their physicochemical characteristics and performance in engine tests. Although these compounds will reduce particulate emissions, their potential environmental impacts are unknown. As a means of characterizing their persistence in environmental media such as soil and

groundwater, we conducted a series of biodegradation tests of DBM and TGME. Benzene and methyl tertiary butyl ether (MTBE) were also tested as reference compounds. Primary degradation of DBM fully occurred within 3 days, while TGME presented a lag phase of approximately 8 days and was not completely degraded by day 28. Benzene primary degradation occurred completely by day 3 and MTBE did not degrade at all. The total mineralized fractions of DBM and TGME achieved constant values as a function of time of ~65% and ~40%, respectively. Transport predictions show that, released to the environment, DBM and TGME would concentrate mostly in soils and waters with minimal impact to air. From an environmental standpoint, these results combined with the transport predictions indicate that DBM is a better choice than TGME as a diesel additive.

Alois Orlita. (Laboratory of Industrial Microbiology, Janáčkova 1110, Otrokovice 765 02, Czech Republic). Microbial biodeterioration of leather and its control: a review. International Biodeterioration & Biodegradation, 53(3) (2004), 157-163.

Hides and leather can be damaged by bacteria, which are mainly responsible for the decomposition of untanned proteins (in raw hides and during soaking), and fungi, which thrive on tanned leathers containing carbohydrates, fats and proteins. A number of fungicides, including 2-(thiocyanomethylthio)benzothiazole, in the wide range of biocidal products now available for preventing defects of biological origin in hides and leathers have been compared in efficacy tests and their penetration, absorption and distribution in tanned leather has been investigated. For protection against moulds it is possible to use a single active ingredient, but it is recommended that a combination of fungicides is used to improve performance by synergistic effects and by broadening the activity spectrum against moulds.

Andrès Y.; Texier A.C.; Le Cloirec P. Rare Earth Elements Removal By Microbial Biosorption: A Review. Environmental Technology, 24(11) (2003), 1367-1375.

This paper reviews published work on the sorption of rare earth elements by microbial biomass. In a first part, the biosorption capacities and the various experimental conditions performed in batch reactor experiments are compared. Secondly, sorption modelling generally used in biosorption studies are described. Thirdly, the microbial cell wall characteristics of the metallic ion binding sites are considered. From these observations it seems that the important functional groups for metallic ion fixation are the carboxyl and the phosphate moieties. Moreover, the competing effect of various ions like aluminium, iron, glutamate, sulphate etc. is described. Finally, some adsorption results of the rare earth elements in dynamic reactors are presented.

Anthony Verdin , Anissa Lounès-Hadj Sahraoui, Roger Durand. (Laboratoire de Mycologie/Phytopathologie/Environnement, Université du Littoral-Côte d'Opale, 17 avenue Blériot, BP 699, Calais Cedex 62228, France). Degradation of benzo[a]pyrene by mitosporic fungi and extracellular oxidative enzymes. International Biodeterioration & Biodegradation, 53(2) (2004), 65-70.

The involvement of extracellular oxidative enzymes (laccase, lignin peroxidase and manganese-dependent peroxidase) in the degradation of benzo[a]pyrene, a high molecular weight polycyclic aromatic hydrocarbon, by three mitosporic fungi (Deuteromycetes) isolated from polluted soils was examined. These fungal strains were found to have different abilities to degrade benzo[a]pyrene: relative degradation percentages per unit biomass for *Trichoderma viride*, *Fusarium solani* and *Fusarium oxysporum* were approximately 39, 17 and 8, respectively. No peroxidase activities were detected in any of the fungal strains. Laccase activities were measured in *F. solani* and *F. oxysporum* cultures, but the specific activities assessed were similar in both whether in the presence or absence of benzo[a]pyrene. Extracellular laccase and peroxidase activities were not induced by benzo[a]pyrene. Moreover, laccase activities of *F. oxysporum*, a poor benzo[a]pyrene

degrader, were fivefold higher than in *F. solani*, a better benzo[a]pyrene degrader. The use of a laccase inhibitor, sodium azide, did not decrease benzo[a]pyrene degradation but the laccase activity was inhibited by 50%. The fact that *T. viride* degrades benzo[a]pyrene more efficiently, without any detectable laccase activity, confirms this result. In conclusion, in these three fungal strains, no apparent correlation between degradation percentage and the tested ligninolytic enzymes production could be shown in our culture conditions.

B.V. Chang, C.H. Yu, S.Y. Yuan. (Department of Microbiology, Soochow University, Shih Lin, Taipei 111, Taiwan). Degradation of nonylphenol by anaerobic microorganisms from river sediment. Chemosphere, 55(4) (2004), 493-500.

We investigated the degradation of nonylphenol monoethoxylate (NP1EO) and nonylphenol (NP) by anaerobic microbes in sediment samples collected at four sites along the Erren River in southern Taiwan. Anaerobic degradation rate constants (k_1) and half-lives ($t_{1/2}$) for NP (2 $\mu\text{g/g}$) ranged from 0.010 to 0.015 1/day and 46.2 to 69.3 days respectively. For NP1EO (2 $\mu\text{g/g}$), the ranges were 0.009–0.014 1/day and 49.5–77.0 days respectively. Degradation rates for NP and NP1EO were enhanced by increasing temperature and inhibited by the addition of acetate, pyruvate, lactate, manganese dioxide, ferric chloride, sodium chloride, heavy metals, and phthalic acid esters. Degradation was also measured under three anaerobic conditions. Results show the high-to-low order of degradation rates to be sulfate-reducing conditions > methanogenic conditions > nitrate-reducing conditions. The results show that sulfate-reducing bacteria, methanogen, and eubacteria are involved in the degradation of NP and NP1EO, with sulfate-reducing bacteria being a major component of the river sediment.

B. V. Chang, C. M. Yang, C. H. Cheng, S. Y. Yuan. (Department of Microbiology, Soochow University, Shih Lin, Taipei 111, Taiwan. Department of Veterinary Medicine, National Taiwan University, Taipei 106, Taiwan). Biodegradation of phthalate esters by two bacteria strains. Chemosphere, 55(4) (2004), 533-538.

In this study two aerobic phthalic acid ester (PAE) degrading bacteria strains, DK4 and O18, were isolated from river sediment and petrochemical sludge, respectively. The two strains were found to rapidly degrade PAE with shorter alkyl-chains such diethyl phthalate (DEP), dipropyl phthalate (DPrP), di-n-butyl phthalate (DBP), benzylbutyl phthalate (BBP) and diphenyl phthalate (DPP) are very easily biodegraded, while PAE with longer alkyl-chains such as dicyclohexyl phthalate (DCP) and dihexyl phthalate (DHP) and di-(2-ethylhexyl) phthalate (DEHP) are poorly degraded. The degradation rates of the eight PAEs were higher for strain DK4 than for strain O18. In the simultaneous presence of strains DK4 and O18, the degradation rates of the eight PAEs examined were enhanced. When the eight PAEs were present simultaneously, degradation rates were also enhanced. We also found that PAE degradation was delayed by the addition of nonylphenol or selected polycyclic aromatic hydrocarbons (PAHs) at a concentration of 1 $\mu\text{g/g}$ in the sediment. The bacteria strains isolated, DK4 and O18, were identified as *Sphigomonas* sp. and *Corynebacterium* sp., respectively.

Benoit Guieysse, Gunilla Viklund, Ann-Charlotte Toes, Bo Mattiasson. (Department of Biotechnology, Lund University, P.O. Box 124, S-221 00, Lund, Sweden). Combined UV-biological degradation of PAHs. Chemosphere, 55(11) (2004), 1493-1499.

The UV-photolysis of PAHs was tested in silicone oil and tetradecane. In most cases, the degradation of a pollutant provided within a mixture was lower than when provided alone due to competitive effects. With the exception of anthracene, the larger pollutants (4- and 5-rings) were always degraded first, proving that UV-treatment preferentially acts on large PAHs and thereby provides a good complement to microbial degradation. UV-photolysis was

also found to be suitable for treatment of soil extract from contaminated soils. The feasibility of UV-biological treatment was demonstrated for the removal of a mixture of phenanthrene and pyrene in silicone oil. UV-irradiation of the silicone oil led to 83% pyrene removal but no phenanthrene photodegradation. Subsequent treatment of the oil in a two-phases partitioning bioreactor (TPPB) system inoculated with *Pseudomonas* sp. was followed by complete phenanthrene biodegradation but no further pyrene removal. Totally, the combined process allowed 92% removal of the PAH mixture. Further work should focus on characterizing the photoproducts formed and studying the influence of the solvent on the photodegradation process.

Benoit Van Aken, Jong Moon Yoon, Jerald L. Schnoor. (Department of Civil and Environmental Engineering, The University of Iowa, Iowa City, Iowa 52242). Biodegradation of Nitro-Substituted Explosives 2,4,6-Trinitrotoluene, Hexahydro-1,3,5-Trinitro-1,3,5-Triazine, and Octahydro-1,3,5,7-Tetranitro-1,3,5-Tetrazocine by a Phytosymbiotic *Methylobacterium* sp. Associated with Poplar Tissues (*Populus deltoides* x *nigra* DN34). Applied and Environmental Microbiology, 70(1) (2004), 508-517.

A pink-pigmented symbiotic bacterium was isolated from hybrid poplar tissues (*Populus deltoides* x *nigra* DN34). The bacterium was identified by 16S and 16S-23S intergenic spacer ribosomal DNA analysis as a *Methylobacterium* sp. (strain BJ001). The isolated bacterium was able to use methanol as the sole source of carbon and energy, which is a specific attribute of the genus *Methylobacterium*. The bacterium in pure culture was shown to degrade the toxic explosives 2,4,6-trinitrotoluene (TNT), hexahydro-1,3,5-trinitro-1,3,5-triazene (RDX), and octahydro-1,3,5,7-tetranitro-1,3,5-tetrazocine (HMX). [U-ring-¹⁴C]TNT (25 mg liter⁻¹) was fully transformed in less than 10 days. Metabolites included the reduction derivatives amino-dinitrotoluenes and diamino-nitrotoluenes. No significant release of ¹⁴CO₂ was recorded from [¹⁴C]TNT. In addition, the isolated methylotroph was shown to transform [U-¹⁴C]RDX (20 mg liter⁻¹) and [U-¹⁴C]HMX (2.5 mg liter⁻¹) in less than 40 days. After 55 days of incubation, 58.0% of initial [¹⁴C]RDX and 61.4% of initial [¹⁴C]HMX were mineralized into ¹⁴CO₂. The radioactivity remaining in solution accounted for 12.8 and 12.7% of initial [¹⁴C]RDX and [¹⁴C]HMX, respectively. Metabolites detected from RDX transformation included a mononitroso RDX derivative and a polar compound tentatively identified as methylenedinitramine. Since members of the genus *Methylobacterium* are distributed in a wide diversity of natural environments and are very often associated with plants, *Methylobacterium* sp. strain BJ001 may be involved in natural attenuation or in situ biodegradation (including phytoremediation) of explosive-contaminated sites.

Bernd Marschner, Karsten Kalbitz. (Department Soil Science/Soil Ecology, Ruhr-University Bochum, D-44780, Bochum, Germany. Department of Soil Ecology, Bayreuth Institute for Terrestrial Ecosystem Research (BITÖK), University of Bayreuth, D-95440, Bayreuth, Germany). Controls of bioavailability and biodegradability of dissolved organic matter in soils. Geoderma, 113(3-4) (2003), 211-235.

In soils, dissolved organic matter (DOM) is probably the most bioavailable fraction of soil organic matter, since all microbial uptake mechanisms require a water environment. Bioavailability describes the potential of microorganisms to interact with DOM. It is a prerequisite for biodegradation and can be restricted, if DOM is present in small pores or within soil aggregates and therefore not accessible for microorganisms. DOM biodegradation is defined as the utilisation of organic compounds by soil microorganisms quantified by the disappearance of DOM or O₂ or by the evolution of CO₂. The controlling factors for DOM biodegradability can be divided into three groups, namely, intrinsic DOM quality parameters, soil and solution parameters and external factors. DOM characteristics that generally

enhance its biodegradability are high contents of carbohydrates, organic acids and proteins for which the hydrophilic neutral fraction seems to be a good estimate. In contrast, aromatic and hydrophobic structures that can also be assessed by UV absorbance decrease DOM biodegradability, either due to their recalcitrance or due to inhibiting effects on enzyme activity. Effects of solution parameters such as Al, Fe, Ca and heavy metal concentrations on DOM biodegradability have been documented in various studies, however with different, sometimes conflicting results. Inhibitory effects of metals are generally attributed to toxicity of the organic complexes or the free metal ions. In contrast, the enhanced degradability observed in the presence of metal ions may be due to flocculation, as larger structures will provide better attachment for microbial colonies. As degradation is dependent on microbial activity, the composition and density of the microbial population used in the degradation studies also influence biodegradation. Site-specific factors, such as vegetation, land use and seasonality of meteorological parameters control DOM composition and soil and soil solution properties and therefore also affect its biodegradability. The major obstacle for a better understanding of the controls of DOM biodegradability is the lack of a standardised methodology or at least systematic comparisons between the large number of methods used to assess DOM biodegradability.

Bethany M. McRae , Timothy M. LaPara and Raymond M. Hozalski. (Department of Civil Engineering, University of Minnesota, 122 Civil Engineering Building, 500 Pillsbury Drive SE, Minneapolis, MN 55455, USA). Biodegradation of haloacetic acids by bacterial enrichment cultures. *Chemosphere*, 55(6) (2004), 915-925.

Haloacetic acids (HAAs) are toxic organic chemicals that are frequently detected in surface waters and in drinking water distribution systems. The aerobic biodegradation of HAAs was investigated in serum bottles containing a single HAA and inoculated with washed microorganisms obtained from enrichment cultures maintained on either monochloroacetic acid (MCAA) or trichloroacetic acid (TCAA) as the sole carbon and energy source. Biodegradation was observed for each of the HAAs tested at concentrations similar to those found in surface waters and in drinking water distribution systems. The MCAA culture was able to degrade both MCAA and monobromoacetic acid (MBAA) with pseudo-first order rate constants of 1.06×10^{-2} and 1.13×10^{-2} l (mg protein)⁻¹ d⁻¹, respectively, for concentrations ranging from 10^{-5} to 2 mM. The pseudo-first order rate constant for TCAA degradation by the TCAA culture was 6.52×10^{-3} l (mg protein)⁻¹ d⁻¹ for concentrations ranging from 5.33×10^{-5} to 0.72 mM. The TCAA culture was also able to degrade MCAA with the rate accelerating as incubation time increased. Experiments with radiolabeled HAAs indicated that the ¹⁴C was primarily converted to ¹⁴CO₂ with minor incorporation into cell biomass. The community structure of the enrichment cultures was analyzed by both cultivation-dependent and cultivation-independent approaches. Denaturing gradient gel electrophoresis (DGGE) of the PCR-amplified 16S rRNA gene fragments showed that each of the two enrichment cultures had multiple bacterial populations, none of which corresponded to HAA-degrading bacteria cultivated on HAA-supplemented agar plates. This research indicates that biodegradation is a potential loss mechanism for HAAs in surface waters and in drinking water distribution systems.

Bill W. Bogan, Wendy R. Sullivan. (Gas Technology Institute, 1700 South Mt. Prospect Road, Des Plaines, IL 60018, USA). Physicochemical soil parameters affecting sequestration and mycobacterial biodegradation of polycyclic aromatic hydrocarbons in soil. *Chemosphere*, 52(10) (2003), 1717-1726.

Six soils, obtained from grasslands and wooded areas in Northeastern Illinois, were physicochemically characterized. Measured parameters included total organic carbon (TOC) content, contents of humic acid, fulvic acid and humin, pore volume and pore size distribution, and chemical makeup of soil organic matter (determined using solid-state ¹³C-

NMR). Moistened, gamma-sterilized soils were spiked with 200 ppm of either phenanthrene or pyrene (including ^{14}C label); following 0, 40, or 120 days of aging, the contaminant-spiked soils were then inoculated with *Mycobacterium austroafricanum* strain GTI-23, and evolution of $^{14}\text{CO}_2$ was assessed over a 28-day period. Results for both phenanthrene and pyrene indicated that increased contact time led to increased sequestration and reduced biodegradation, and that TOC content was the most important parameter governing these processes. One soil, although only tested with phenanthrene, showed significantly lower-than-expected sequestration (higher-than-expected mineralization) after 40 days of aging, despite a very high TOC value (>24%). Because the level of sequestration in this soil was proportional to the others after 120 days of aging, this implies some difference in the temporal progression of sequestration in this soil, although not in its final result. The primary distinguishing feature of this soil was its considerably elevated fulvic acid content. Further experiments showed that addition of exogenous fulvic acid to a soil with very low endogenous humic acids/fulvic acids content greatly enhanced pyrene mineralization by *M. austroafricanum*. Extractabilities of 13 three- to six-ring coal tar PAHs in n-butanol from the six soils after 120 days of sequestration were strongly TOC-dependent; however, there was no discernible correlation between n-butanol extractability and mycobacterial PAH mineralization.

Bo Gejlsbjerg, Torben Madsen, Trine Thorup Andersen. (DHI Water and Environment, Department of Ecotoxicology--Agern Allé 11, DK 2970, Hørsholm, Denmark). Comparison of biodegradation of surfactants in soils and sludge-soil mixtures by use of ^{14}C -labelled compounds and automated respirometry. *Chemosphere*, 50(3) (2003), 321-331.

The biodegradability of dodecyl benzene sulphonate (LAS), nonylphenol-di-ethoxylate (NP2EO) and tridecyl-tetra-ethoxylate (LAE) in soil was examined by use of ^{14}C experiments at two concentrations (10 and 400 mg/kg). Increasing the concentration of test chemical from 10 to 400 mg/kg resulted in a decrease in the relative maximum mineralization rate and an increase in the estimated lag times of a factor of approximately 3.5. In sludge-amended soil, the highest expected environmental concentration (just after sludge application) will be around 10 mg/kg for linear alkylbenzene sulphonate (LAS), while the concentration of NP2EO and linear alcohol ethoxylates (LAE) will be much lower. However, when using a respirometric method it is necessary to use a higher concentration of test substance in order to detect biodegradation. In our experiment, amendment with anaerobically digested sludge resulted in a decrease in the mineralization of LAS, NP2EO and LAE for all soils. Respirometric experiments were carried out at 400 mg/kg and could be used for estimation of biodegradation potential of LAS, NP2EO and LAE in soil and sludge-amended soil. For LAS, the results obtained from the respirometric experiments were similar to the results obtained in the ^{14}C experiments, whereas NP2EO and LAE showed a faster degradation in the respirometric experiments.

Brajesh K. Singh, Allan Walker, J. Alun W. Morgan, Denis J. Wright. (Horticulture Research International, Wellesbourne, Warwick CV35 9EF, Department of Biological Sciences, Imperial College at Silwood Park, Ascot, Berkshire SL5 7PY, United Kingdom). Role of Soil pH in the Development of Enhanced Biodegradation of Fenamiphos. *Applied and Environmental Microbiology*, 69(12) (2003), 7035-7043.

Repeated treatment with fenamiphos (ethyl 4-methylthio-m-tolyl isopropylphosphoramidate) resulted in enhanced biodegradation of this nematicide in two United Kingdom soils with a high pH (7.7). In contrast, degradation of fenamiphos was slow in three acidic United Kingdom soils (pH 4.7 to 6.7), and repeated treatments did not result in enhanced biodegradation. Rapid degradation of fenamiphos was observed in two

Australian soils (pH 6.7 to 6.8) in which it was no longer biologically active against plant nematodes. Enhanced degrading capability was readily transferred from Australian soil to United Kingdom soils, but only those with a high pH were able to maintain this capability for extended periods of time. This result was confirmed by fingerprinting bacterial communities by 16S rRNA gene profiling of extracted DNA. Only United Kingdom soils with a high pH retained bacterial DNA bands originating from the fenamiphos-degrading Australian soil. A degrading consortium was enriched from the Australian soil that utilized fenamiphos as a sole source of carbon. The 16S rRNA banding pattern (determined by denaturing gradient gel electrophoresis) from the isolated consortium migrated to the same position as the bands from the Australian soil and those from the enhanced United Kingdom soils in which the Australian soil had been added. When the bands from the consortium and the soil were sequenced and compared they showed between 97 and 100% sequence identity, confirming that these groups of bacteria were involved in degrading fenamiphos in the soils. The sequences obtained showed similarity to those from the genera *Pseudomonas*, *Flavobacterium*, and *Caulobacter*. In the Australian soils, two different degradative pathways operated simultaneously: fenamiphos was converted to fenamiphos sulfoxide (FSO), which was hydrolyzed to the corresponding phenol (FSO-OH) or was hydrolyzed directly to fenamiphos phenol. In the United Kingdom soils in which enhanced degradation had been induced, fenamiphos was oxidized to FSO and then hydrolyzed to FSO-OH, but direct conversion to fenamiphos phenol did not occur.

Buenaventurada P. Calabia, Yutaka Tokiwa. (Department of Science and Technology, Industrial Technology Development Institute, Bicutan, Taguig, Metro Manila 1604, Philippines. National Institute of Advanced Industrial Science and Technology (AIST), Tsukuba Central 6, 1-1-1 Higashi, Tsukuba, Ibaraki 305-8566, Japan). Microbial degradation of poly(d-3-hydroxybutyrate) by a new thermophilic *Streptomyces* isolate. *Biotechnology Letters*, 26(1) (2004), 15-19.

A new thermophilic microorganism capable of degrading poly(d-3-hydroxybutyrate) (PHB) was isolated from soil. A phylogenetic analysis based on 16S rDNA sequences indicated that the new isolate belongs to genus *Streptomyces*. PHB film and powder were completely degraded after 6 and 3 d cultivation, respectively at 50 °C. Scanning micrographs showed adherence of the microbial cells to the entire film surface, indicating that biodegradation occurs by colonization of the PHB surface. The film was degraded both by microbial attack and by the action of an extracellular enzyme secreted by the microorganism. The strain can also degrade poly(ethylene succinate), poly(ester carbonate), polycaprolactone and poly(butylene succinate), but to a lesser extent.

Carla A. Nicholson and Babu Z. Fathepure. (Department of Microbiology and Molecular Genetics, Oklahoma State University, Stillwater, Oklahoma 74078-3020). Biodegradation of Benzene by Halophilic and Halotolerant Bacteria under Aerobic Conditions. *Applied and Environmental Microbiology*, 70(2) (2004), 1222-1225.

A highly enriched halophilic culture was established with benzene as the sole carbon source by using a brine soil obtained from an oil production facility in Oklahoma. The enrichment completely degraded benzene, toluene, ethylbenzene, and xylenes within 1 to 2 weeks. Also, [¹⁴C]benzene was converted to ¹⁴CO₂, suggesting the culture's ability to mineralize benzene. Community structure analysis revealed that *Marinobacter* spp. were the dominant members of the enrichment.

Chunlong Zhang, Joseph B. Hughes. (Department of Environmental Sciences, University of Houston-Clear Lake, Houston, TX 77058-1098, USA. Department of Civil and Environmental Engineering, Rice University, Houston, TX 77005, USA). Biodegradation pathways of hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX) by

***Clostridium acetobutylicum* cell-free extract. Chemosphere, 50(5) (2003), 665-671.**

Hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX), a military high explosive, is becoming an increasingly important pollutant in the US. The cleanup of RDX-contaminated soil and groundwater has been a serious challenge due to its recalcitrance in the environment. This study was conducted to determine the biodegradation kinetics of RDX by crude cell extract of *Clostridium acetobutylicum* (ATCC 824), and to examine whether this bacterium will carry out reductive transformation pathways similar to the transformation of 2,4,6-trinitrotoluene (TNT), 2,4- and 2,6-dinitrotoluenes (DNTs) we have reported previously. Batch studies on the anaerobic transformation of RDX were conducted in serum bottles with U-ring-¹⁴C-RDX. RDX and its transformation products were quantified by HPLC and qualified by LC/MS interfaced to two soft ionization techniques--an atmospheric pressure ionization and an electron spray ionization (API-ES). Results demonstrated that *C. acetobutylicum* is capable of transforming RDX with H₂ as the electron donor. The transformation followed a zero-order kinetics and the rates increased with increasing H₂. RDX was transformed into several polar intermediates that could not be separated by reverse-phase HPLC and its molecular ions were unstable under the condition of commonly used electron impact detector. Using a polar and water immiscible solvent (ethyl acetate) and the softer MS ionization techniques, mass spectroscopy detected the presence of several RDX derivatives including mononitroso-, monohydroxylamino-, mononitrosomonohydroxylamino-, monoamino-, diamino-, and triamino-compounds. The presence of hydroxylamino compounds is analogous to the transformation of TNT and DNTs we elucidated previously.

Cynthia Davis, Todd Cort, Dongping Dai, Tissa H. Illangasekare, Junko Munakata-Marr. (URS Corporation, 9801 Westheimer, Suite 500, Houston, TX 77042, USA Environmental Science and Engineering Division, Colorado School of Mines, Golden, CO 80401, USA. Environmental Science and Engineering Division, Colorado School of Mines, Golden, CO 80401, USA. Cameron-Cole, LLC, 5777 Central Avenue, Suite 100, Boulder, CO 80301, USA). Effects of Heterogeneity and Experimental Scale on the Biodegradation of Diesel. Biodegradation, 14(6) (2003), 373-384.

Biodegradation of petroleum hydrocarbon contamination is a common method for remediating soils and groundwater. Due to complexities with field-scale studies, biodegradation rates are typically evaluated at the bench-scale in laboratory studies. However, important field conditions can be difficult to mimic in the laboratory. This study investigates three scaling factors that can impact laboratory biodegradation rates and that are frequently unaccounted for in typical laboratory experimental procedures. These factors are soil heterogeneity, morphology of petroleum hydrocarbon non-aqueous phase liquids (NAPLs) and soil moisture distribution. The effects of these factors on the biodegradation rate of diesel NAPL is tested under a variety of experimental procedures from well-mixed batch studies to four-foot static soil columns. The results indicate that a high degree of variability results from even small-scale heterogeneities. In addition, it appears that as the experimental scale increases, the measured biodegradation rates slow. The results indicate that diesel biodegradation rates derived from small-scale experiments are not necessarily representative of field-scale biodegradation rates.

D. Moldes, S. Rodríguez Couto, C. Cameselle, M. A. Sanromán. (Department of Chemical Engineering. University of Vigo, E-36200 Vigo, Spain). Study of the degradation of dyes by MnP of *Phanerochaete chrysosporium* produced in a fixed-bed bioreactor. Chemosphere, 51(4) (2003), 295-303.

The production of ligninolytic enzymes by the fungus *Phanerochaete chrysosporium* in a fixed-bed tubular bioreactor, filled with cubes of nylon sponge, operating in semi-solid-state conditions, was studied. Maximum individual manganese-dependent peroxidase (MnP) and

lignin peroxidase (LiP) activities of 1293 and 225 U/l were detected. The in vitro decolourisation of two structurally different dyes (Poly R-478, crystal violet) by the extracellular liquid obtained in the above-mentioned bioreactor was monitored in order to determine its degrading capability. The concentration of some compounds (sodium malonate, manganese sulphate) from the reaction mixture was optimised in order to maximise the decolourisation levels. A percentage of Poly R-478 decolourisation of 24% after 15 min of dye incubation was achieved. On the other hand, a methodology for a long treatment of these dyes based on the continuous addition of MnP enzyme and H₂O₂ was developed. Moreover, this enzymatic treatment was compared with a photochemical decolourisation process. The former allowed to maintain the degradation rate almost constant for a long time, resulting in a decolourisation percentage of 70% and 30% for crystal violet and Poly R-478, respectively, after 2 h of treatment. As for the latter, it was not able to degrade Poly R-478, whereas crystal violet reached a degradation of 40% in 2 h.

Daljit Singh Arora, Mukesh Chander and Paramjit Kaur Gill. (Department of Microbiology, Microbial Technology Laboratory, Guru Nanak Dev University, Amritsar 143 005, India). Involvement of lignin peroxidase, manganese peroxidase and laccase in degradation and selective ligninolysis of wheat straw. International Biodeterioration & Biodegradation, 50(2) (2002), 115-120.

Some white-rot fungi have been studied to evaluate their potential to degrade wheat straw with specific reference to their ligninolytic ability and associated enzymes, viz. lignin peroxidase, manganese peroxidase, and laccase. The data have been compared with the much studied *Phanerochaete chrysosporium*. In the present study, *Daedalea flavida* and two of the *Phlebia* spp. were found to be capable of degrading lignin selectively and hence hold better prospects in various biotechnological applications than *P. chrysosporium*. *Phlebia radiata* and *P. floridensis* were the best producers of manganese peroxidase and laccase, respectively, whereas *P. chrysosporium* was best for lignin peroxidase.

Darryl P. Arfsten, Dennis T. Burton, Daniel J. Fisher, John Callahan, Cody L. Wilson, Kenneth R. Still, Barry J. Spargo. (Naval Health Research Center Detachment Environmental Health Effects Laboratory (Toxicology)--NHRC/TD, Bldg. 433, 2612 5th Street, Wright-Patterson AFB, OH 45433-7903, USA. University of Maryland, Wye Research and Education Center, Queenstown, MD 21658, USA. Naval Research Laboratory, Washington, DC 20375, USA. Navy Environmental and Preventive Medicine Unit 2, 1887 Powhatan Street, Norfolk, VA 23511, USA). Assessment of the aquatic and terrestrial toxicity of five biodegradable polymers. Environmental Research, 94(2) (2004), 198-210.

Radiofrequency countermeasures (i.e., chaff) may be released by fighter jets during tactical countermeasures training. Chaff cartridges, pistons, and endcaps (i.e., chaff dispenser materials), all currently made of styrene, are also released into the environment. Accumulation of chaff dispenser materials in the environment is a concern of the Department of Defense. The US Navy is exploring the possibility of constructing degradable chaff dispenser components made of biodegradable polymers. Five polymers are being considered. Degradability and toxicity tests are two of several criteria being used to evaluate various available biodegradable options. Dissolution products from four of five polymers being considered were toxic to aquatic organisms with LC50s/LOELs ranging between 1.24 and 731.30 mg total organic concentration/L. Supernatant from dissolving a 90:10 polyester amide/polyvinyl alcohol copolymer in water for 24 h inhibited shoot growth of *Brassica rappa* and *Lepidium sativum*. Since our results were obtained using fractions of saturated degradable polymer solutions (1 or 10 g/L), we conclude that the tested degradable polymers were of low toxicity to the seven aquatic organisms and two terrestrial plant species used in our assays. However, our characterization of the toxicity of these

degradable polymers may not be applicable to all species or environmental situations. Information gained from these studies will be used for making decisions on which polymers should be used in the engineering of environmentally friendly chaff dispenser cartridges, pistons, and endcaps.

Diane Fournier, Annamaria Halasz, Jim Spain, Ronald J. Spanggord, Jeffrey C. Bottaro, Jalal Hawari. (Biotechnology Research Institute, National Research Council of Canada, Montreal, Quebec H4P 2R2, Canada, U.S. Air Force Research Laboratory, Tyndall Air Force Base, Florida 32403,² SRI International, Menlo Park, California 94025). Biodegradation of the Hexahydro-1,3,5-Trinitro-1,3,5-Triazine Ring Cleavage Product 4-Nitro-2,4-Diazabutanal by *Phanerochaete chrysosporium*. *Applied and Environmental Microbiology*, 70(2) (2004), 1123-1128.

Initial denitration of hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX) by *Rhodococcus* sp. strain DN22 produces CO₂ and the dead-end product 4-nitro-2,4-diazabutanal (NDAB), OHCNHCH₂NHNO₂, in high yield. Here we describe experiments to determine the biodegradability of NDAB in liquid culture and soils containing *Phanerochaete chrysosporium*. A soil sample taken from an ammunition plant contained RDX (342 μmol kg⁻¹), HMX (octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine; 3,057 μmol kg⁻¹), MNX (hexahydro-1-nitroso-3,5-dinitro-1,3,5-triazine; 155 μmol kg⁻¹), and traces of NDAB (3.8 μmol kg⁻¹). The detection of the last in real soil provided the first experimental evidence for the occurrence of natural attenuation that involved ring cleavage of RDX. When we incubated the soil with strain DN22, both RDX and MNX (but not HMX) degraded and produced NDAB (388 ± 22 μmol kg⁻¹) in 5 days. Subsequent incubation of the soil with the fungus led to the removal of NDAB, with the liberation of nitrous oxide (N₂O). In cultures with the fungus alone NDAB degraded to give a stoichiometric amount of N₂O. To determine C stoichiometry, we first generated [¹⁴C]NDAB in situ by incubating [¹⁴C]RDX with strain DN22, followed by incubation with the fungus. The production of ¹⁴CO₂ increased from 30 (DN22 only) to 76% (fungus). Experiments with pure enzymes revealed that manganese-dependent peroxidase rather than lignin peroxidase was responsible for NDAB degradation. The detection of NDAB in contaminated soil and its effective mineralization by the fungus *P. chrysosporium* may constitute the basis for the development of bioremediation technologies.

E. G. Gregoricha, M. H. Beareb, U. Stoklasa, P. St-Georges. (Agriculture Canada, Central Experimental Farm, Ottawa, Canada K1A 0C6. New Zealand Institute for Crop and Food Research, Christchurch, New Zealand). Biodegradability of soluble organic matter in maize-cropped soils. *Geoderma*, 113(3-4) (2003), 237-252.

Soluble organic matter and its biodegradability are important in relation to soil nutrient fluxes, carbon (C) sequestration, and water quality. The objective of this study was to evaluate the quantity and biodegradability of soluble organic matter in soil under monoculture maize or maize-soybean rotation, and different amendments: manure, inorganic fertilizer, or no amendment. We characterized organic matter extracted in cold and hot (80 °C) water by using a bioassay involving incubation of the extracts (after inoculation with soil microflora) at 35 °C for 42 days. Soluble organic C and organic and mineral nitrogen (N) were monitored during the incubation. Extracts of whole soil (0–15 cm depth) and three water-stable aggregate size classes (>1000, 1000–250, and 250–50 μm) were subjected to the bioassay in order to evaluate the physical disposition and kinetics of soluble organic matter. Hot water-soluble C accounted for about 70% of the total (cold+hot) water-soluble organic matter. Organic N was the major form of N in the extracts; it comprised 61–83% of the total N extracted with cold water and 87–97% of the total N extracted with hot water. The quantity of soluble organic matter that was biodegradable was related to the extraction procedure and the management history of the soil. The proportion of soluble C and organic N that was biodegradable was greatest in hot water

extracts and greater in manured than nonmanured soils. During the bioassay, proportionately greater amounts of organic N were metabolized relative to organic C, so that the C:N ratios of the extracts widened as the incubation progressed, indicating that the soluble organic matter was rich in labile organic N. The rate at which soluble organic matter decomposed (i.e., turnover) was not related to the type of input. Decomposition data fitted a double exponential decay model, suggesting that the soluble organic matter comprised two fractions: a rapidly decomposable fraction (containing 29–36% of the total soluble C) with a turnover time of <1 day, and a slowly decomposable fraction with a turnover time of about 80 days. The concentration of soluble organic matter in soil aggregates was related to aggregate size; the concentration was greatest in the largest aggregates and it decreased with decreasing aggregate size. Soluble organic C extracted from aggregates also comprised two kinetically discrete pools. The turnover time (i.e., mean residence time) of each pool was not affected by amendment, crop rotation, or aggregate size. However, the slowly decomposable organic C pool was larger in microaggregates (<250 μm) than in macroaggregates, whereas the rapidly decomposable pool was larger in macroaggregates. We conclude that water-soluble organic matter in maize-cropped soils comprises two kinetically defined fractions and is readily decomposable due to its high content of hydrophilic, labile N-rich compounds.

Elizalde-González M.P., Peláez-Cid A.A. Removal of Textile Dyes from Aqueous Solutions by Adsorption on Biodegradable Wastes. *Environmental Technology*, 24(7) (2003), 821-829.

The adsorption of the textile dyes Basic Blue 41, Aniline Blue, Reactive Black 5 and Mariposa Blue was studied from model solutions prepared in distilled water in the concentration range up to 2000 mg l⁻¹. Biodegradable wastes were used and are proposed here as adsorbents due to their abundance and inexpensiveness in the authors' country. The influence of the particle size, pH, presence of an electrolyte in the dye solution, adsorbent activation and competition in ternary dyes mixtures was determined in this study. Adsorption capacity of the adsorbents designated as HOMAP and POMAP with particle diameter less than 3 mm for the Basic Blue 41 dye was found to be 164 and 77 mg g⁻¹, respectively. The efficiency of the prepared adsorbents for removing basic, acid and reactive dyes with a concentration of 400 mg l⁻¹ was examined. A comparison of the adsorption capacity and desorption feasibility of the adsorbents was made. The basic dye was adsorbed with efficiencies varying from 87 - 98%, the reactive dye was removed in the range 60 - 98% and the acid dye, 60 - 70% by three of the four adsorbents tested in this study. The determination of adsorption equilibrium concentrations was followed by HPLC with diode array detection.

Fabio Fava, Silvia Gentilucci, Giulio Zanaroli. (DICASM, Faculty of Engineering, University of Bologna, viale Risorgimento 2, I-40136, Bologna, Italy). Anaerobic biodegradation of weathered polychlorinated biphenyls (PCBs) in contaminated sediments of Porto Marghera (Venice Lagoon, Italy). *Chemosphere*, 53(2) (2003), 101-109.

The biodegradation of weathered polychlorinated biphenyls (PCBs) (mono and di-chlorinated biphenyls along with PCBs partially ascribed to Aroclor 1242 and 1254) occurring at 1.5–2.5 mg/kg in three different sediments collected from the Porto Marghera contaminated area of Venice Lagoon (Italy) was reported in this study. Strictly anaerobic, slurry microcosms consisting of sediments suspended (at 25% v/v) in a marine salt medium, lagoon water or lagoon water supplemented with NaHCO₃ and Na₂S were developed and monitored for PCB transformation, sulfate consumption and methane (CH₄) production for 6 months. A marked depletion of highly chlorinated biphenyls along with the accumulation of low-chlorinated, often ortho-substituted biphenyls was observed in the biologically active microcosms, where a remarkable consumption of sulfate and/or a significant production of CH₄ were also

detected. Notably, a more extensive PCB transformation was observed in the microcosms developed with site water (both without or with NaHCO₃ plus Na₂S), where both the initial concentration of sulfate and sulfate consumption were five fold-higher than in the corresponding microcosms with salt medium. These data indicate that weathered PCBs of the three contaminated sediments of Porto Marghera utilized in this study can undergo reductive dechlorination, probably mediated by indigenous sulfate-reducing and/or methanogenic bacteria.

Farinazleen Mohamad Ghazali , Raja Noor Zaliha Abdul Rahman, Abu Bakar Salleh and Mahiran Basri. (Department of Biochemistry & Microbiology, Enzyme and Microbial Technology Research, Faculty of Science and Environmental Studies, Universiti Putra Malaysia, UPM Serdang, Selangor 43400, Malaysia). Biodegradation of hydrocarbons in soil by microbial consortium. International Biodeterioration & Biodegradation, 54(1) (2004), 61-67.

The bioremediation of hydrocarbon in contaminated soils by mixed cultures of hydrocarbon-degrading bacteria was investigated. The mixtures or consortia of bacteria, denoted as Consortium 1 and Consortium 2 consisted of 3 and 6 bacterial strains, respectively. Bacterial strains used in this study were from the Center for Research in Enzymes and Microbiology (CREAM) collection of strains, at Universiti Putra Malaysia, and were isolated from hydrocarbon-contaminated soil samples by enrichments on either crude oil or individual hydrocarbons as the sole carbon source. The strains were selected based on the criteria that they were able to display good growth in crude oil, individual hydrocarbon compounds or both. Their ability to degrade hydrocarbon contamination in the environment was investigated using soil samples that were contaminated with diesel, crude oil or engine oil. Consortium 2, which consisted of 6 bacterial strains, was more efficient at removing the medium- and long-chain alkanes in the diesel-contaminated soil compared to Consortium 1. Further, Consortium 2 could effectively remove the medium- and long-chain alkanes in the engine oil such that the alkanes were undetectable after a 30-day incubation period. Consortium 2 consisted predominantly of *Bacillus* and *Pseudomonas* spp.

H. Kang, S.Y. Hwang, Y.M. Kim, E. Kim, Y.-S. Kim, S.-K. Kim, S.W. Kim, C.E. Cerniglia, K.L. Shuttleworth, and G.J. Zylstra. Degradation of phenanthrene and naphthalene by a Burkholderia species strain. Canadian Journal of Microbiology, 49(2) (2003), 139-144.

Burkholderia sp. TNFYE-5 was isolated from soil for the ability to grow on phenanthrene as sole carbon and energy source. Unlike most other phenanthrene-degrading bacteria, TNFYE-5 was unable to grow on naphthalene. Growth substrate range experiments coupled with the ring-cleavage enzyme assay data suggest that TNFYE-5 initially metabolizes phenanthrene to 1-hydroxy-2-naphthoate with subsequent degradation through the phthalate and protocatechuate and \square -ketoadipate pathway. A metabolite in the degradation of naphthalene by TNFYE-5 was isolated by high-pressure liquid chromatography (HPLC) and was identified as salicylate by UV-visible spectral and gas chromatography – mass spectrometry analyses. Thus, the inability to degrade salicylate is apparently one major reason for the incapability of TNFYE-5 to grow on naphthalene.

I.J. Díaz-Ramírez, H. Ramírez-Saad, M. Gutiérrez-Rojas, E. Favela-Torres. Biodegradation of Maya crude oil fractions by bacterial strains and a defined mixed culture isolated from *Cyperus laxus* rhizosphere soil in a contaminated site. Canadian Journal of Microbiology, 49(12) (2003), 755-761.

Ten bacterial strains were isolated by enrichment culture, using as carbon sources either aliphatics or an aromatic–polar mixture. Oxygen uptake rate was used as a criterion to determine culture transfer timing at each enrichment stage. Biodegradation of aliphatics (10

000 mg L⁻¹) and an aromatic-polar mixture (5000 mg L⁻¹, 2:1) was evaluated for each of the bacterial strains and for a defined culture made up with a standardized mixture of the isolated strains. Degradation of total hydrocarbons (10 000 mg L⁻¹) was also determined for the defined mixed culture. Five bacterial strains were able to degrade more than 50% of the aliphatic fraction. The most extensive biodegradation (74%) was obtained with strain Bs 9A, while strains Ps 2AP and UAM 10AP were able to degrade up to 15% of the aromatic-polar mixture. The defined mixed culture degraded 47% of the aliphatics and 6% of the aromatic-polar mixture. The defined mixed culture was able to degrade about 40% of the aliphatic fraction and 26% of the aromatic fraction when grown in the presence of total hydrocarbons, while these microorganisms did not consume the polar hydrocarbons fraction. The proposed strategy that combines enrichment culture together with oxygen uptake rate allowed the isolation of bacterial strains that are able to degrade specific hydrocarbons fractions at high consumption rates.

Isabel Sierra, José Luis Valera, M. Luisa Marina, and Fernando Laborda. (Centro de Tecnología de los Alimentos y Servicios, Biosanitarios, Universidad de Alcalá, 28871 Alcalá de Henares, Madrid, Spain. Departamento de Química Analítica, Facultad de Química, Universidad de Alcalá, 28871 Alcalá de Henares, Madrid, Spain. Departamento de Microbiología y Parasitología, Facultad de Farmacia, Universidad de Alcalá, 28871 Alcalá de Henares, Madrid, Spain). Study of the biodegradation process of polychlorinated biphenyls in liquid medium and soil by a new isolated aerobic bacterium (*Janibacter* sp.). *Chemosphere*, 53(6) (2003), 609-618.

We have isolated and characterised a novel aerobic bacterial strain, designated MS3-02, belonging to the genus *Janibacter* sp. The capability of this new strain to degrade polychlorinated biphenyls (PCBs) in a commercial mixture (Aroclor 1242) in liquid medium and in soil (sterile and non sterile soil), under laboratory scale, has been evaluated. MS3-02 was isolated from the soil around of an incinerator, located in the east of Madrid (Spain). Gas-chromatographic analysis showed that MS3-02 was able to reduce most peaks observed in the chromatogram between 70% and 100% after seven days of incubation in a culture mineral medium containing yeast extract, but without the addition of biphenyl. The presence of biphenyl in the culture medium decreased the rate of PCB degradation by this bacterium. Comparing the performance of the MS3-02 in liquid culture medium and in soil, degradation was less efficient in sterile soil and still less efficient in non sterile soil. Under the best conditions (sterile soil and 20 weeks of incubation) MS3-02 was able to reduce, between 50% and 100%, nine of the main gas-chromatographic peaks in Aroclor 1242.

Iveta Řezníčková, Jaromír Hoffmann, Karel Komárek. (Faculty of Technology, Department of Environmental Technology and Chemistry, Tomas Bata University in Zlín, náměstí T.G. Masaryka 275, 762 72 Zlín, Czech Republic. Faculty of Chemical Technology, University of Pardubice, náměstí Čs. legií 565, 532 10 Pardubice, Czech Republic). Biodegradation of technical mixtures of oxyethylenated aliphatic alcohols in an aqueous environment. *Chemosphere*, 48(1) (2002), 83-87.

A study of the biodegradability of nonionogenic surfactants of oxyethylenated n-butyl, isobutyl, n-pentyl and isopentyl alcohols was conducted by modified static test according to International Standard Organization method 9888. Degradation was evaluated by changes in concentrations of individual oligomers, ethylene glycols, carboxylic and dicarboxylic acids. Good biological degradability was found, with a "primary" degradability of 75-98% in 10 days. Obtained results, in agreement with our previous work, proved the dominant proportion of degradative scission in the hydrophile part of surfactant molecule. During biological degradation no significant cumulation of mono- to polyethylene glycols appeared

neither that of mono- or dicarboxylic acids, and degradation of these components was not a rate-determining degradation factor.

Jadwiga Szostak-Kotowa. (Department of Microbiology, Cracow University of Economics str. Rakowicka 27, Katedra Mikrobiologii, Akademia Ekonomiczna w Krakowie, 31-510, Kraków, Poland). Biodeterioration of textiles. International Biodeterioration & Biodegradation, 53(3) (2004), 165-170.

Textiles, particularly those composed of natural organic fibres such as cotton, linen, wool, etc., are readily attacked by microorganisms. Most synthetic fabrics are not readily subject to extensive biodeterioration, but some processing and finishing agents are susceptible to microbial spoilage. Microorganisms can affect all stages of textile processing and storage, with fungi being the most important microorganisms in textile biodeterioration processes. Microbial growth on a textile causes loss of strength and elongation, discolouration and changes in appearance. They follow changes in oxidation state, degree of polymerization and breakdown of molecular structure. There are two main ways of textile protection-- control of the environmental physical conditions and treatments with biocides.

Jae Jun Jeong, Ji Hyun Kim, Chi-Kyung Kim, Ingyu Hwang and Kyoung Lee. (Department of Microbiology, Changwon National University. Kyongnam 661-773. Korea, Department of Microbiology, Chungbuk National University, Cheongju 361-736. Korea. School of Agricultural Biotechnology, Seoul National University. Seoul 151-742. Korea). 3- and 4-alkylphenol degradation pathway Pseudomonas sp. strain KL28: genetic organization of the lap gene cluster and substrate specificities of phenol hydroxylase and 2,3-dioxygenase. Microbiology, 149 (2003), 3265-3277.

The enzymes and genes responsible for the catabolism of higher alkylphenols have not been characterized in aerobic bacteria. *Pseudomonas* sp. strain KL28 can utilize a wide range of alkylphenols, which include the 4-n-alkylphenols (C1-C5). The genes, designated as lap (for fang-chain alky/phenols), encoding enzymes for the catabolic pathway were cloned from chromosomal DNA and sequenced. The lap genes are located in a 13-2 kb region with 14 ORFs in the order lapRBKLMNOPCEHIFG and with the same transcriptional orientation. The lapR gene is transcribed independently and encodes a member of the XylR/DmpR positive transcriptional regulators. LapB, the first gene in the lap operon, encodes catechol 2,3-dioxygenase (C230). The lapKLMNOP and lapCEHIFG genes encode a multicomponent phenol hydroxylase (mPH) and enzymes that degrade derivatives of 2-hydroxymuconic semialdehyde (HMS) to TCA cycle intermediates, respectively. The PlapB promoter contains motifs at positions -24(GG) and -12(GC) which are typically found in σ 54-dependent promoters. A promoter assay using a PlapB::gfp transcriptional fusion plasmid showed that lapB promoter activity is inducible and that it responds to a wide range of (alkyl)phenols. The structural genes encoding enzymes required for this catabolism are similar (42-69%) to those encoded on a catabolic pVI150 plasmid from an archetypal phenol degrader, *Pseudomonas* sp. CF600. However, the lap locus does not include genes encoding HMS hydrolase and ferredoxin. The latter is known to be functionally associated with C230 for use of 4-alkylcatechols as substrates. The arrangement of the lap catabolic genes is not commonly found in other meta-cleavage operons. Substrate specificity studies show that mPH preferentially oxidizes 3- and 4-alkylphenols to 4-alkylcatechols. C230 preferentially oxidizes 4-alkylcatechols via proximal (2,3) cleavage. This indicates that these two key enzymes have unique substrate preferences and lead to the establishment of the initial steps of the lap pathway in strain KL28.

Jaka Widada, Hideaki Nojiri, Takako Yoshida, Hiroshi Habe, Toshio Omori. (Biotechnology Research Center, The University of Tokyo, 1-1-1 Yayoi, Bunkyo-ku, Tokyo 113-8657, Japan. Laboratory of Soil and Environmental Microbiology, Department of Soil Science, Faculty of Agriculture, Gadjah Mada University,

Bulaksumur, Yogyakarta 55281, Indonesia). Enhanced degradation of carbazole and 2,3-dichlorodibenzo-*p*-dioxin in soils by *Pseudomonas resinovorans* strain CA10. *Chemosphere*, 49(5) (2002), 485-491.

We studied the degradation of carbazole (CAR) and 2,3-dichlorodibenzo-*p*-dioxin (2,3-DCDD) in soils inoculated with carbazole- and dioxin-degrader *Pseudomonas resinovorans* strain CA10. By using Tn5-based transposon delivery systems, this bacterium was chromosomally marked with a tandem green fluorescent protein (*gfp*) gene. Real-time competitive PCR and direct counting using the (*gfp*) marker were employed to monitor the total number of carbazole 1,9a-dioxygenase gene (*carAa*) and survival of CA10 cells in the soil and soil slurry microcosms. Bioaugmentation studies indicated that the survival of the marked CA10 cells in soil microcosms was strongly influenced by pH and organic matter. While the number of the marked CA10 cells decreased rapidly in pH 6 with low organic matter, a high cell density was maintained in pH 7.3 with 2.5% organic matters up to 21 days after inoculation. In pH 7.3 soil, the period needed for complete degradation of CAR (100 $\mu\text{g kg}^{-1}$) was markedly shortened from 21 to 7 days by the inoculation with the CA10 cells. Single inoculation of CA10 cells into the soil slurry system of 2,3-DCDD-contaminated soil enhanced the degradation of 2,3-DCDD from 25.0% to 37.0%. In this system, the population density of CA10 cells and the total number of *carAa* gene were maintained up to 14 days after inoculation. By repeated inoculation (every 2 days) with CA10 cells each at a density of 10^9 CFU g^{-1} of soil, almost all of the 2,3-DCDD (1 $\mu\text{g kg}^{-1}$) was degraded within 14 days. Results of these experiments suggest that *P. resinovorans* strain CA10 may be an important resource for bioremediation of CAR and chlorinated dibenzo-*p*-dioxin in contaminated soils.

Jens Dittmann, Wolfgang Heyser and Heike Bücking. (Center for Environmental Research and Technology (UFT), Applied Botany, Plant Anatomy and Physiology, University of Bremen, D-28359, Bremen, Germany). Biodegradation of aromatic compounds by white rot and ectomycorrhizal fungal species and the accumulation of chlorinated benzoic acid in ectomycorrhizal pine seedlings. *Chemosphere*, 49(3) (2002), 297-306.

The capability of different white rot (WR, *Heterobasidion annosum*, *Phanerochaete chrysosporium*, *Trametes versicolor*) and ectomycorrhizal (ECM, *Paxillus involutus*, *Suillus bovinus*) fungal species to degrade different aromatic compounds and the absorption of 3-chlorobenzoic acid (3-CBA) by ECM pine seedlings was examined. The effect of aromatic compounds on the fungal biomass development varied considerably and depended on (a) the compound, (b) the external concentration, and (c) the fungal species. The highest effect on the fungal biomass development was observed for 3-CBA. Generally the tolerance of WR fungi against aromatic compounds was higher than that of the biotrophic fungal species. The capability of different fungi to degrade aromatic substances varied between the species but not generally between biotrophic and saprotrophic fungi. The highest degradation capability for aromatic compounds was detected for *T. versicolor* and *H. annosum*, whereas for *Phanerochaete chrysosporium* and the ECM fungi lower degradation rates were found. However, *Paxillus involutus* and *S. bovinus* showed comparable degradation rates at low concentrations of benzoic acid and 4-hydroxybenzoic acid. In contrast to liquid cultures, where no biodegradation of 3-CBA by *S. bovinus* was observed, mycorrhizal pines inoculated with *S. bovinus* showed a low capability to remove 3-CBA from soil substrates. Additional X-ray microanalytical investigations showed, that 3-CBA supplied to mycorrhizal plants was accumulated in the root cell cytoplasm and is translocated across the endodermis to the shoot of mycorrhizal pine seedlings.

Ji-Guang Gu, Yanzhen Fan, Ji-Dong Gu. (Key Laboratory of Terrestrial Ecological Process, Shenyang Institute of Applied Ecology, Chinese Academy of Sciences,

Shenyang, Liaoning 10015, PR China. Laboratory of Environmental Toxicology, Department of Ecology & Biodiversity, The University of Hong Kong, Pokfulam Road, Hong Kong SAR, PR China. Environmental and Molecular Microbiology, South China Sea Institute of Oceanography, Chinese Academy of Sciences, 164 West Xingang Road, Guangzhou 510301, Guangdong, PR China). Biodegradability of Atrazine, Cyanazine and Dicamba under methanogenic condition in three soils of China. *Chemosphere*, 52(9) (2003), 1515-1521.

Persistence and degradation of the herbicides Atrazine, Cyanazine and Dicamba were measured in laboratory microcosms incubated under methanogenic condition using three soils of China. Results showed that Atrazine was more resistant to degradation than Cyanazine and Dicamba for the 300 days of incubation. Between 30% and 40% of the initially introduced chemicals were found to be not recoverable through solvent extraction of the incubated soils. Our results also indicated that the half-life of these herbicides in the three soils generally followed: Atrazine > Cyanazine > Dicamba. Biodegradation of Cyanazine and Dicamba was further substantiated by establishing enrichment cultures in which the degradation of the respective herbicides could be accelerated by the microorganisms. Our results suggest that biodegradation of xenobiotics can be established through enrichment culture transfer technique and non-extractability of chemicals should be taken into account in evaluation of chemicals' fate and risk.

Joanita Coelho, C. U. Rivonkar, N. S. Bhavesh., M. Jothi & U. M. X. Sangodkar. Biosurfactants production by the quinoline degrading marine bacterium *Pseudomonas* sp. strain GU 104, and its effect on the metabolism of green mussel *Perna viridis* L. *Indian Journal of Marine Sciences*, 32(3) (2003), 202-207.

Biosurfactants produced by bacteria in marine ecosystems are involved in the degradation of hydrocarbons. In the present study, large-scale production of biosurfactants was demonstrated in a quinoline degrading marine bacterium *Pseudomonas* sp. strain GU 104. Studies were also carried out in experimental set ups to understand the effect of biosurfactants, along with the metabolites of quinoline, on the physiology of the green mussel *Perna viridis*. Acetylcholinesterase (AChE), lactate dehydrogenase (LDH), phenoloxidase and α -amylase activities from specific organs were analysed. The findings of the present study indicate that biosurfactant, as well as quinoline intermediates, produced by *Pseudomonas* sp. strain GU 104 do not have a significant effect on the physiology of *Perna viridis*.

Jukka Ahtiainen, Miia Aalto, Piia Pessala. (Finnish Environment Institute, P.O. Box 140, FIN-00251, Helsinki, Finland). Biodegradation of chemicals in a standardized test and in environmental conditions. *Chemosphere*, 51(6) (2003), 529-537.

The estimation of biodegradation rates is an important source of uncertainty in chemical risk assessment. The existing OECD tests for ready biodegradability have been developed to devise screening methods to determine whether a chemical is potentially easily biodegradable, rather than to predict the actual rate, of biodegradation in the environment. However, risk assessment needs degradation rates. In practice these rates are often estimated (default values) from ready biodegradability tests. These tests have many compromising arbitrary features compared to the situation in the real environment. One important difference is the concentration of the chemical. In wastewater treatment or in the environment many chemicals are present at ng l^{-1} to $\mu\text{g l}^{-1}$ levels whereas in the tests the concentrations exceed 10–400 mg carbon per litre. These different concentrations of the chemical will lead to different growth kinetics and hence different biodegradation rates. At high concentrations the chemical, if it is degradable, can serve as a primary substrate and competent microorganisms will grow exponentially, resulting in a sigmoid biodegradation curve. At low environmental concentrations the chemical does not serve as a primary

substrate, and therefore does not support significant growth of the degraders, and the substrate has a linear biodegradation rate. In this study the biodegradation rates of two reference chemicals, aniline and 4-chloroaniline, were compared in a standard method and in more realistic conditions at low concentrations, using ^{14}C -labelled substances and different sources of inocula. Biomass evolution during the tests was monitored by adenosine triphosphate measurement and also on the basis of the residual ^{14}C -activity in the particulate matter. The results partly support the thesis that low concentrations lead to different biodegradation kinetics compared to the concentrations used in the standard tests. Furthermore the biodegradation rates of the chemicals studied, particularly of 4-chloroaniline, in Finnish natural waters appeared to be lower than those reported in some other countries.

Jung-Hyun Lee, Seong-Young Jung, Sang-Jin Kim. (Microbiology Laboratory, Korea Ocean Research & Development Institute, Ansan P.O. Box 29, Seoul 425-600, Korea). Specific detection of an oil-degrading bacterium, *Corynebacterium* sp. IC10, in sand microcosms by PCR using species-specific primers based on 16S rRNA gene sequences. *Biotechnology Letters*, 23(21) (2001), 1741-1748.

A species-specific PCR technique to detect an oil-degrading bacterium, *Corynebacterium* sp. IC10, released into sand microcosms is described. PCR primers, specific to strain IC10, were designed based on 16S rRNA gene sequences and tested against both closely and distantly related bacterial strains using four primer combinations involving two forward and two reverse primers. Two sets of them were specific to the strain IC10 and *Corynebacterium variabilis* and one set was selected for further analysis. The PCR amplification was able to detect 1 pg template DNA of strain IC10 and 1.2×10^4 c.f.u. of IC10 ml wet sand-1 in the presence of 3×10^8 *Escherichia coli* cells. In non-sterile sand microcosms seeded with the strain IC10, the sensitivity of detection decreased to 9.6×10^5 c.f.u. ml wet sand-1. The detection sensitivity thus depends on the complexity of background heterogeneous DNA of environmental samples. The assay is suitable for detection of *Corynebacterium* sp. IC10 in laboratory microcosms, however, cross reaction with non-oil degrading coryneforms may prohibit its use in uncharacterized systems.

K. Kalbitz, J. Schmerwitz, D. Schwesig, E. Matzner. (Department of Soil Ecology, Bayreuth Institute for Terrestrial Ecosystem Research (BITÖK), University of Bayreuth, D-95440, Bayreuth, Germany). Biodegradation of soil-derived dissolved organic matter as related to its properties. *Geoderma*, 113(3-4) (2003), 273-291.

Quantifying the contribution of dissolved organic matter (DOM) to C sequestration in soils requires knowledge about extent and rate of its biodegradation. Since degradation experiments are time consuming, estimating the biodegradability of DOM by more easily measurable properties seems valuable. Our goal was therefore to investigate the biodegradation of DOM of different origin and to relate its extent and rate to properties such as UV absorbance, synchronous and emission scan fluorescence, XAD-8 sorption chromatography and $^1\text{H-NMR}$ spectroscopy. We extracted DOM from 13 different samples (maize straw, forest floors, peats, agricultural soils) and carried out a 90-day liquid incubation experiment. DOM biodegradation was quantified by CO_2 evolution. Rapidly and slowly mineralizable portions of dissolved organic carbon (DOC) as a measure of labile and stable DOC and the respective mineralization rate constants and half-lives were calculated by a double exponential model. The extent and rate of DOM biodegradation from less humified organic material (straw, litter and fermentation layers of forest floors) were high resulting in 61–93% of DOC being mineralized. The labile fraction comprised 59–88% of total DOC. DOM extracted from agricultural soils was of an intermediate biodegradability with a CO_2 evolution comprising 17–32% of total DOC. Labile DOC represented 14–25% of total DOC. DOM extracted from peats and Oa forest floor layers was relatively stable

(mineralization of 4–9% of total DOC, labile DOC: 3–6%). The half-life of the labile DOC pool was short (2–5 days), whereas that of the stable DOC pool ranged from 0.2 years (DOM from less humified material) to 8.6 years (DOM from the Oa layer under spruce). Extent and rate of DOM biodegradation were closely but nonlinearly related to DOM properties. Solutions exceeding a threshold value of UV absorbance, aromaticity, XAD-8 adsorbable C or of humification indices derived from fluorescence spectra or having carbohydrate contents below a certain level were stable against biodegradation. Relatively simple methods like UV spectroscopy, XAD-8 fractionation and fluorescence emission spectroscopy were suitable to estimate the biodegradation of DOM.

K. Padma Dorothy, B. Satyanarayana, C. Kalavati, A. V. Raman, F. Dehairs. (Department of Zoology, Andhra University, Visakhapatnam – 530003 (AP), India. Vrije Universiteit Brussel, Department of Analytical Chemistry (ANCH), Pleinlaan 2, B-1050 Brussels, Belgium). Protozoa associated with leaf litter degradation in Coringa mangrove forest, Kakinada Bay, east coast of India. Indian Journal of Marine Sciences, 32(1) (2003), 45-51.

Observations (1995-'96) on mangrove leaf litter revealed a variety of microorganisms dominated by bacteria (5 types), 12 species of flagellates, 2 sarcodines, 17 ciliates, 2 suctorids and 2 sessile ciliates besides several diatoms, nematodes and nauplii. Overall, bacteria outnumbered (4.59 others constituting 80-90% of the population followed by flagellates (4.8%), ciliates (4.4%) and, sessile ciliates (0.2%). *Chromulina* sp., *Spumella socialis* and *Euglena acus* (flagellates), *Cyclidium* sp., *Prorodon* sp., *Euplotoides aediculatus* and *Zoothamnium* sp. (ciliates) were relatively dominant (mean density 4,331 individuals l⁻¹) in the litter collected from *Avicennia* plot. Flagellates, *Astasia* sp., *Heteronema* sp. and *Paranema* sp. and, ciliates, *Prorodon* sp., *Holosticha* sp. and *E. aediculatus* were, however, more common in *Excoecaria* (mean density 3719 individuals l⁻¹). In situ experiments on leaf decay showed that the entire process lasted 12-18 days in summer and 26-32 days during monsoon. Bacteria were the first to settle, followed by nanoflagellates (2-20 μm), microciliates (20-100 μm), macrociliates (100-200 μm) and sessile ciliates. Nematodes indicated culmination. Bacterial (mean) bio-mass registered highest value (6043 ± 10-3 mgC g⁻¹) within 24 hours but decreased (3.1 ± 10-6 mgC g⁻¹) by day-3 to 5. Mean flagellate biomass peaked (32.6 mgC g⁻¹) by day-2 and microciliates (92 mgC g⁻¹) by day-5 in summer and (47mgC g⁻¹) by day-24 during monsoon. Macrociliates registered highest biomass (168.4mgC g⁻¹) by day-6 in summer but lagged behind until day-26 to day-30 (154mgC g⁻¹) during monsoon. A distinct prey predator relationship, direct dependence of ciliate species on nanoflagellate and bacterial populations as well as, a well marked microbial community succession were evident.

Kari T. Steffen, Annele Hatakka, Martin Hofrichter. (Division of Microbiology, Department of Applied Chemistry and Microbiology, University of Helsinki, FIN-00014 Helsinki, Finland, 1 Unit of Environmental Biotechnology, International Graduate School Zittau, D-02763 Zittau, Germany). Degradation of Benzo[a]pyrene by the Litter-Decomposing Basidiomycete *Stropharia coronilla*: Role of Manganese Peroxidase. Applied and Environmental Microbiology, 69(7) (2003), 3957-3964.

The litter-decomposing basidiomycete *Stropharia coronilla*, which preferably colonizes grasslands, was found to be capable of metabolizing and mineralizing benzo[a]pyrene (BaP) in liquid culture. Manganese(II) ions (Mn²⁺) supplied at a concentration of 200 μM stimulated considerably both the conversion and the mineralization of BaP; the fungus metabolized and mineralized about four and twelve times, respectively, more of the BaP in the presence of supplemental Mn²⁺ than in the basal medium. This stimulating effect could be attributed to the ligninolytic enzyme manganese peroxidase (MnP), whose activity

increased after the addition of Mn²⁺. Crude and purified MnP from *S. coronilla* oxidized BaP efficiently in a cell-free reaction mixture (in vitro), a process which was enhanced by the surfactant Tween 80. Thus, 100 mg of BaP liter⁻¹ was converted in an in vitro reaction solution containing 1 U of MnP ml⁻¹ within 24 h. A clear indication was found that BaP-1,6-quinone was formed as a transient metabolite, which disappeared over the further course of the reaction. The treatment of a mixture of 16 different polycyclic aromatic hydrocarbons (PAHs) selected by the U.S. Environmental Protection Agency as model standards for PAH analysis (total concentration, 320 mg liter⁻¹) with MnP resulted in concentration decreases of 10 to 100% for the individual compounds, and again the stimulating effect of Tween 80 was observed. Probably due to their lower ionization potentials, poorly bioavailable, high-molecular-mass PAHs such as BaP, benzo(g,h,i)perylene, and indeno(1,2,3-c,d)pyrene were converted to larger extents than low-molecular-mass ones (e.g., phenanthrene and fluoranthene).

Kazunari Sei¹, Yoshiro Sugimoto¹, Kazuhiro Mori¹, Hideaki Maki, Tetsuro Kohno. Monitoring of alkane-degrading bacteria in a sea-water microcosm during crude oil degradation by polymerase chain reaction based on alkane-catabolic genes. Environmental Microbiology, 5(6) (2003), 517.

Behaviour of microbial populations responsible for degrading n-alkanes, a major component of crude oil, was monitored during crude oil degradation in a sea-water microcosm by both traditional colony culturing and molecular techniques. A DNA extraction method applicable to crude oil-amended sea-water samples was developed to obtain DNA applicable to most probable number (MPN) polymerase chain reaction (PCR). The population of alkane-degrading bacteria responsible for degradation of n-alkanes in a crude oil-amended microcosm altered, so that shorter alkanes were degraded first by alkane-degrading bacteria possessing alkane hydroxylase genes from group I (Kohno et al., 2002, Microb Environ 17: 114-121) and longer ones afterwards by those possessing alkane hydroxylase genes from group II. Thus, the degradation mechanism of the n-alkanes can be clarified during crude oil degradation. Application of the method of detecting different types of alkane-catabolic genes, as shown in the present study, enabled bacterial groups preferring alkanes of either shorter or longer chain lengths to be enumerated selectively.

Krassimira Hristova, Binyam Gebreyesus, Douglas Mackay, and Kate M. Scow. (Department of Land, Air, and Water Resources, University of California, Davis, California). Naturally Occurring Bacteria Similar to the Methyl tert-Butyl Ether (MTBE)-Degrading Strain PM1 Are Present in MTBE-Contaminated Groundwater. Applied and Environmental Microbiology, 69(5) (2003), 2616-2623.

Methyl tert-butyl ether (MTBE) is a widespread groundwater contaminant that does not respond well to conventional treatment technologies. Growing evidence indicates that microbial communities indigenous to groundwater can degrade MTBE under aerobic and anaerobic conditions. Although pure cultures of microorganisms able to degrade or cometabolize MTBE have been reported, to date the specific organisms responsible for MTBE degradation in various field studies have not been identified. We report that DNA sequences almost identical (99% homology) to those of strain PM1, originally isolated from a biofilter in southern California, are naturally occurring in an MTBE-polluted aquifer in Vandenberg Air Force Base (VAFB), Lompoc, California. Cell densities of native PM1 (measured by TaqMan quantitative PCR) in VAFB groundwater samples ranged from below the detection limit (in anaerobic sites) to 10³ to 10⁴ cells/ml (in oxygen-amended sites). In groundwater from anaerobic or aerobic sites incubated in microcosms spiked with 10 µg of MTBE/liter, densities of native PM1 increased to approximately 10⁵ cells/ml. Native PM1 densities also increased during incubation of VAFB sediments during MTBE degradation. In controlled field plots amended with oxygen, artificially increasing the MTBE concentration was followed by

an increase in the in situ native PM1 cell density. This is the first reported relationship between in situ MTBE biodegradation and densities of MTBE-degrading bacteria by quantitative molecular methods.

Lars Elsgaard, Giulio Pojana, Tommaso Miraval, Jørgen Eriksen, Antonio Marcomini. (Department of Crop Physiology and Soil Science, Danish Institute of Agricultural Sciences, Research Center Foulum, P.O. Box 50, DK-8830, Tjele, Denmark. Department of Environmental Sciences, University of Venice, Calle Larga S. Marta 2137, I-31023, Venice, Italy). Biodegradation of linear alkylbenzene sulfonates in sulfate-leached soil mesocosms. *Chemosphere*, 50(7) (2003), 929-937.

Aromatic sulfonates ($R-SO_3^-$) can be used as sulfur sources by sulfate-starved bacteria in laboratory cultures and the corresponding phenols are excreted from the cells. The present study was conducted to demonstrate whether such desulfonation reactions also occur in sulfate-leached agricultural soil, where desulfonation of organic sulfur compounds may have agronomic importance as a S source for plants. Xenobiotic linear alkylbenzene sulfonates (LAS) were added to nominal concentrations of 0, 10 and 100 mg kg⁻¹ dry weight in a sandy soil that was depleted in sulfate by leaching the soil with water (sulfate depletion, ~75%). The soil was incubated at 20 °C in duplicate 3-dm³ mesocosms for 8 weeks. Primary degradation of LAS was rapid with half-lives of 1–4 days. Sulfophenylcarboxylates were identified and quantified as intermediates, whereas linear alkylphenols (the expected primary desulfonation products) were not detected by high-pressure liquid chromatography coupled with both fluorescence and electrospray ionization-mass spectrometry. Thus, LAS was used by the bacteria as a source of energy and carbon, rather than as a source of sulfur. Measurements of soil pH, fluorescein diacetate (FDA) hydrolysis and arylsulfatase activity showed that stable microbial conditions prevailed in the soil mesocosms. FDA hydrolysis (a measure of total microbial activity) was transiently inhibited at the highest LAS concentrations. Arylsulfatase activity (i.e., hydrolysis of aromatic sulfate esters) was not significantly affected by the soil incubation, although arylsulfatases may be upregulated in sulfate-starved bacteria. However, an increased production of arylsulfatase may be difficult to detect due to the background of extracellular arylsulfatases stabilised in the soil. Therefore, the present data does not exclude a regulatory response to sulfate depletion by the soil microorganisms. However, the importance of desulfonation reactions in natural environments still needs to be demonstrated.

Leonóra Száraz, Judit Becznerb. (Szent István University, Faculty of Food Science, Villányi út 29-33, 1118, Budapest, Hungary. Department of Microbiology, Central Food Research Institute, Herman Ottó út 15, 1022, Budapest, Hungary). Optimization processes of a CO₂ measurement set-up for assessing biodegradability of polymers. *International Biodeterioration & Biodegradation*, 52(2) (2003), 115-125.

A method originating in the measurement of biological activity of soils was adapted and optimized to assess biodegradability of polymers in controlled composting conditions. 2 g sample embedded in 80 g compost aerated at 70 mlmin⁻¹ was found as an optimal set-up to reach an adequate amount of determined CO₂.

Lepo J.E., Cripe C.R., Kavanaugh J.L., Zhang S., Norton G.P. The Effect of Amount of Crude Oil on Extent of its Biodegradation in Open Water- and Sandy Beach-Laboratory Simulations. *Environmental Technology*, 24(10) (2003), 1291-1302.

We examined the biodegradation of varying amounts of artificially weathered Alaskan North Slope crude oil in laboratory microcosm test systems that use natural seawater and simulate spills in open water and on sandy beaches. The model bioremediation treatment

consisted of periodic applications of marine bacteria, selected to degrade n-alkanes and a range of aromatic compounds, suspended in a salts solution that supplied inorganic nitrogen and phosphorous. Beach microcosms dosed with low and high oiling lost an average of 22.5% and 11.3% oil weight, respectively. Open-water microcosms dosed with high and low oiling lost 19.1% and 2.9% oil weight, respectively. Thus, the lower doses of oil were more efficiently degraded. The model bioremediation treatment also affected a greater number of selected analytical endpoints in the lower-oil-dose than higher-dose experiments and the former showed more substantial degradation of recalcitrant components. Above a certain threshold oil concentration, bioremediation did not effectively remove oil. Below this threshold the distinction between active bioremediation treatment and intrinsic biodegradation of the controls was less prominent; i.e., fewer of the oil components were statistically depleted by remediation treatment relative to controls. Furthermore, the oil dose range over which bioremediation was realized in these systems occurred at very low oiling levels. Thus, under the environmental conditions simulated in these microcosms, the effectiveness of bioremediation peaked over a rather narrow low-dose oiling range.

Lourenço N.D., Novais J.M., Pinheiro H.M. Analysis of Secondary Metabolite Fate During Anaerobic-Aerobic Azo Dye Biodegradation in a Sequential Batch Reactor. Environmental Technology, 24(6) (2003), 679-686.

A great number of the reported examples of azo dye biodegradation comprise two main steps, the reductive cleavage of the azo bond under anaerobic conditions and the subsequent aerobic mineralization of the produced aromatic amines. Based on this possible metabolism a Sequencing Batch Reactor was chosen to study biological color removal from simulated cotton textile effluents containing a reactive azo dye. In previous studies high color removal levels of the azo dye Remazol Brilliant Violet 5R were achieved (up to 90% with an initial dye concentration of 100 mg l⁻¹) during the anaerobic phase of Sequencing Batch Reactor operation. However, HPLC analyses revealed that the aromatic amines formed in the anaerobic phase were not mineralized during the subsequent aerobic phase. In an attempt to promote the aerobic biodegradation of these aromatic amines three different approaches were tested, the increase of the relative duration of the aerobic phase, the increase of the hydraulic retention time through the decrease of the daily fill flow and finally the increase of the dye/carbon source concentration ratio through the decrease of the fed volumetric organic load. The two aromatic amines directly resulting from azo bond reduction were detected by HPLC analysis. However, a third metabolite with significant peak area was also detected with a time profile suggesting an equilibrium with one of the aromatic amines. In spite of the conversions occurring between metabolites during the cycles of the tested approaches, no effective biodegradation of these metabolites was observed during the experimental period of over 810 days.

Luis A. Rios-Hernandez, Lisa M. Gieg, Joseph M. Suflita. (Institute for Energy and the Environment and Department of Botany and Microbiology, University of Oklahoma, Norman, Oklahoma 73019). Biodegradation of an Alicyclic Hydrocarbon by a Sulfate-Reducing Enrichment from a Gas Condensate-Contaminated Aquifer. Applied and Environmental Microbiology, 69(1) (2003), 434-443.

We used ethylcyclopentane (ECP) as a model alicyclic hydrocarbon and investigated its metabolism by a sulfate-reducing bacterial enrichment obtained from a gas condensate-contaminated aquifer. The enrichment coupled the consumption of ECP with the stoichiometrically expected amount of sulfate reduced. During ECP biodegradation, we observed the transient accumulation of metabolite peaks by gas chromatography-mass spectrometry, three of which had identical mass spectrometry profiles. Mass-spectral similarities to analogous authentic standards allowed us to identify these metabolites as

ethylcyclopentylsuccinic acids, ethylcyclopentylpropionic acid, ethylcyclopentylcarboxylic acid, and ethylsuccinic acid. Based on these findings, we propose a pathway for the degradation of this alicyclic hydrocarbon. Furthermore, a putative metabolite similar to ethylcyclopentylsuccinic acid was also found in samples of contaminated groundwater from the aquifer. However, no such finding was evident for samples collected from wells located upgradient of the gas condensate spill. Microbial community analysis of the ECP-degrading enrichment by denaturing gradient gel electrophoresis revealed the presence of at least three different organisms using universal eubacterial primers targeting 550 bp of the 16S rRNA gene. Based on sequence analysis, these organisms are phylogenetically related to the genera *Syntrophobacter* and *Desulfotomaculum* as well as a member of the *Cytophaga-Flexibacter-Bacteroides* group. The evidence suggests that alicyclic hydrocarbons such as ECP can be anaerobically activated by the addition to the double bond of fumarate to form alkylsuccinate derivatives under sulfate-reducing conditions and that the reaction occurs in the laboratory and in hydrocarbon-impacted environments.

M. Cerón-Rivera, M. M. Dávila-Jiménez, M. P. Elizalde-González. (Centro de Química, Instituto de Ciencias, Universidad Autónoma de Puebla, Apdo. Postal J-55, Puebla, Pue. 72571, Mexico. Facultad de Ciencias Químicas, Universidad Autónoma de Puebla, Apdo. Postal J-55, Puebla, Pue. 72571, Mexico). Degradation of the textile dyes Basic yellow 28 and Reactive black 5 using diamond and metal alloys electrodes. *Chemosphere*, 55(1) (2004), 1-10.

Basic yellow 28 (SLY) and Reactive black 5 (CBWB), which are respectively methine and sulfoazo textile dyes were individually exposed to electrochemical treatment using diamond-aluminium-, copper- and iron-zinc alloy electrodes. The generated current was registered with time during electrolysis of the dye solutions and the color variation and the formation of degradation products were followed using HPLC with diode array detection. Four different electrodic materials were tested by applying different potentials in the range -1.0 to -2.5 V and presented 95% color removal and COD removal of up to 65-67% in the case of CBWB dye solution treated with the copper and iron electrodes. Efficiency was enhanced with stirring and flow in relation to the stationary regime. The kinetic parameter reaction rate was used to establish the effect of flow, potential, electrode nature and pH. The formation and characterization of the precipitate formed under certain conditions is reported and discussed.

M. Fürhacker, A. Pressl, R. Allabashi. (Department for Sanitary Engineering and Water Pollution Control, Institute for Water Provision, Water Ecology and Waste Management, University of Agricultural Sciences, Vienna A-1190, Muthgasse 18, Austria). Aerobic biodegradability of methyldiethanolamine (MDEA) used in natural gas sweetening plants in batch tests and continuous flow experiments. *Chemosphere*, 52(10) (2003), 1743-1748.

Mixtures of different amines including tertiary amines (methyldiethanolamine, MDEA) are commonly used for the removal of CO₂ from gas mixtures or in gas sweetening processes for the extraction of CO₂ and H₂S. The absorber solutions used can be released into the industrial waste water due to continuous substitution of degraded MDEA, periodically cleaning processes or an accidental spill. In this study, the aerobic biodegradability of MDEA was investigated in a standardised batch test and a continuous flow experiment (40 l/d). The results of the batch test indicated that the MDEA-solution was non-biodegradable during the test period of 28 days, whereas the continuous flow experiments showed biodegradation of more than 96% based on TOC-measurements. This was probably due to the adaptation of the microorganisms to this particular waste water contamination during continuous flow experiment.

M. Trigui, S. Pulvin, P. Poupin, and D. Thomas. Biodegradation of cyclic amines by a *Pseudomonas* strain involves an amine mono-oxygenase. *Canadian Journal of Microbiology*, 49(3) (2003), 181-188.

Pseudomonas putida O1G3 catalyzes the degradation of pyrrolidine and piperidine. This strain can use these compounds as the sole source of carbon, nitrogen, and energy. When the cyclic amines were used as the growth substrates, the synthesis of a soluble heme amine mono-oxygenase was induced in this bacteria. This observation was confirmed by spectrophotometric analysis and specific inhibitor. This mono-oxygenase is a NADH-dependent enzyme and catalyzes the cleavage of the C—N bond of the pyrrolidine and piperidine ring by a mechanism similar to a N dealkylation. This reaction could be followed by ring cleavage to form α -aminobutyraldehyde oxidized to α -aminobutyrate. Further investigations to purify the heme-containing mono-oxygenase are in progress.

Mark J. Hopkins, Hans N. Englyst, Sandra Macfarlane, Elizabeth Furrie, George T. Macfarlane, Andrew J. McBain. (MRC Microbiology and Gut Biology Group, University of Dundee, Dundee. Englyst Carbohydrates, Research & Services Ltd., Southampton. School of Pharmacy and Pharmaceutical Sciences, University of Manchester, Manchester, United Kingdom). Degradation of Cross-Linked and Non-Cross-Linked Arabinoxylans by the Intestinal Microbiota in Children. *Applied and Environmental Microbiology*, 69(11) (2003), 6354-6360.

In humans, nonstarch polysaccharides (NSP), such as arabinoxylans (AX), are not digested in the upper gut and provide fermentable carbon sources for bacteria growing in the large bowel. Despite the ubiquity of AX in nature, the microbiologic and physiologic consequences of AX digestion in the gut are poorly understood. In this study, we investigated the breakdown of ferulic acid-cross-linked AX (AXF) and non-cross-linked AX in children's intestinal microbiotas, using starch as a readily fermentable polysaccharide for comparative purposes. The experiments were performed using pH-controlled fermentation vessels under anaerobic conditions. The results demonstrated that there was variation in the metabolism of these polysaccharides by colonic microbiotas. AX was always degraded more slowly than starch, while ferulic acid cross-linking reduced the rate of AX fermentation, as shown by fermentation product measurements. Starch digestion was associated with significant acetate and butyrate production, whereas AX breakdown resulted in increased propionate formation. In general, the presence of fermentable carbohydrate significantly increased the total anaerobe counts and eubacterial rRNA concentrations ($P < 0.01$), while non-cross-linked AX digestion was principally associated with increased viable counts of *Bacteroides fragilis* group organisms, which was supported by increases in *Bacteroides*-*Porphyromonas*-*Prevotella* group rRNA ($P < 0.01$). Starch was considerably more bifidogenic than AX in these fermentations. In conclusion, in this study we found that the effects of AX and AXF on the microbial ecology and metabolism of intestinal microbiotas are similar in children and adults.

Mayra A. Laraa, Antonio J. Rodríguez-Malaverb, Orlando J. Rojas, Otón Holmquistc, Aura M. González, Johnny Bullóna, Nancy Peñalozab, Elisa Araujoa. (Lab. FIRP, Escuela de Ingeniería Química, Facultad de Ingeniería, Universidad de Los Andes, Mérida 5101, Venezuela. Lab. Bioquímica Adaptativa, Facultad de Medicina, Universidad de Los Andes, Mérida 5101, Venezuela. Lab. Patología Forestal, Facultad de Ciencias Forestales y Ambientales, Universidad de Los Andes, Mérida 5101, Venezuela). Black liquor lignin biodegradation by *Trametes elegans*. *International Biodeterioration & Biodegradation*, 52(2) (2003), 93-95.

The white rot fungus *Trametes elegans* was used for direct treatment of spent black liquor from pulping processes with the aim to degrade solubilized lignin which is the primary organic by-product from the chemical digestion of lignocellulosic raw materials. The

enzymatic activity of *T. elegans* and the resulting degradation was confirmed by UV/VIS spectrometric and size-exclusion chromatography (SEC) measurements on incubated and control samples of both industrial and synthetic black liquors. Opposing polymerization and depolymerization reactions occurred after treatment of the black liquor with *T. elegans* in liquid media. It is noteworthy that these effects, which originate from ligninolytic enzyme systems, occur after the direct treatment of the black liquor even if no nutrients are added. The effect of the suspension pH and incubation time on the lignolytic action of *T. elegans* is also presented.

Meltem Urgun-Demirtas, Krishna R. Pagilla, Benjamin C. Stark, Dale Webster. (Department of Chemical and Environmental Engineering, Illinois Institute of Technology Chicago, IL 60616, USA. Department of Biological, Chemical and Physical Sciences, Illinois Institute of Technology Chicago, IL 60616, USA). Biodegradation of 2-Chlorobenzoate by Recombinant *Burkholderia Cepacia* Expressing *Vitreoscilla* Hemoglobin Under Variable Levels of Oxygen Availability. Biodegradation, 14(5) (2003), 357-365.

The influence of bacterial hemoglobin, VHb, on dechlorination and degradation of 2-chlorobenzoate (2-CBA) by recombinant *Burkholderia* sp. under variable oxygen availability with an initial dissolved oxygen concentration of 0.27 mM-0.72 mM was investigated in batch and continuous culture. Ability to express VHb was provided to recombinant *Burkholderia* by transformation with the VHb gene, *vgb*, on plasmid pSC160. 100% of 0.5 mM CBA was degraded in cultures with 85% and 70% of total volume as headspace air in closed reactors by both wild type and recombinant *Burkholderia*. The recombinant cultures were able to dechlorinate and degrade 100% of the 2-CBA in less than 48 hours at 30 °C compared to more than 120 hours for wild type cultures. The rate and extent of CBA degradation by recombinant cultures with 40% of total volume as headspace air was higher than those achieved by wild type cells at the end of the 168 hours of incubation period, 98 and 73%, respectively. The chloride released: CBA degraded molar ratio for cultures with 40% of total volume headspace air was nearly stoichiometric (molar ratio = 1.0) for recombinant strains, whereas it was non-stoichiometric (molar ratio = 0.24) for wild type cells. The results suggest a suicidal *meta*-pathway for wild type cells and a complete dechlorination and degradation pathway for recombinant cells under hypoxic conditions. The degradation and dechlorination ability of both types of cells was also investigated in continuous reactor studies by varying the dilution rate under hypoxic conditions. Regarding potential of the recombinant strain for 2-CBA degradation in either open ecosystems or closed bioreactor bioremediation systems, the stability of the plasmid containing *vgb* in the recombinant cells was also studied; the plasmid was 100% stable at 0.025 h⁻¹ dilution rate (~1.7 d hydraulic retention time), even after one month.

Merja Itävaara, Sari Karjomaa, Johan-Fredrik Selin. (VTT Biotechnology, Tietotie 2, P.O. Box 1500, FIN-02044 VTT, Finland. Fortum Oil and Gas Oy, Technology Centre, P.O. Box 310, FIN-06101 Porvoo, Finland). Biodegradation of polylactide in aerobic and anaerobic thermophilic conditions. Chemosphere, 46(6) (2002), 879-885.

Biodegradable polymers are designed to resist a number of environmental factors during use, but to be biodegradable under disposal conditions. The biodegradation of polylactide (PLLA) was studied at different elevated temperatures in both aerobic and anaerobic, aquatic and solid state conditions. In the aerobic aquatic headspace test the mineralisation of PLLA was very slow at room temperature, but faster under thermophilic conditions. The clear effect of temperature on the biodegradability of PLLA in the aquatic tests indicates that its polymer structure has to be hydrolysed before microorganisms can utilise it as a nutrient source. At similar elevated temperatures, the biodegradation of PLLA was much faster in

anaerobic solid state conditions than in aerobic aquatic conditions. The behaviour of PLLA in the natural composting process was similar to that in the aquatic biodegradation tests, biodegradation starting only after the beginning of the thermophilic phase. These results indicate that PLLA can be considered as a compostable material, being stable during use at mesophilic temperatures, but degrading rapidly during waste disposal in compost or anaerobic treatment facilities.

Michael F. Coughlin, Brian K. Kinkle, Paul L. Bishop. (DiverseyLever, 3630 East Kemper Road, Cincinnati, OH 45241, USA. Department of Biological Sciences, University of Cincinnati, Cincinnati, OH 45221-0006, USA. Department of Civil and Environmental Engineering, University of Cincinnati, Cincinnati, OH 45221-0071, USA). Degradation of acid orange 7 in an aerobic biofilm. *Chemosphere*, 46(1) (2002), 11-19.

A stable microbial biofilm community capable of completely mineralizing the azo dye acid orange 7 (AO7) was established in a laboratory scale rotating drum bioreactor (RDBR) using waste liquor from a sewage treatment plant. A broad range of environmental conditions including pH (5.8–8.2), nitrification (0.0–4.0 mM nitrite), and aeration (0.2–6.2 mg O₂ l⁻¹) were evaluated for their effects on the biodegradation of AO7. Furthermore the biofilm maintained its biodegradative ability for over a year while the effects of these environmental conditions were evaluated. Reduction of the azo bond followed by degradation of the resulting aromatic amine appears to be the mechanism by which this dye is biodegraded. Complete loss of color, sulfanilic acid, and chemical oxygen demand (COD) indicate that AO7 is mineralized. To our knowledge this is the first reported occurrence of a sulfonated phenylazonaphthol dye being completely mineralized under aerobic conditions. Two bacterial strains (1CX and SAD4i) originally isolated from the RDBR were able to mineralize, in co-culture, up to 90% of added AO7. During mineralization of AO7, strain 1CX reduces the azo bond under aerobic conditions and consumes the resulting cleavage product 1-amino-2-naphthol. Strain SAD4i consumes the other cleavage product, sulfanilic acid. The ability of the RDBR biofilm to aerobically mineralize an azo dye without exogenous carbon and nitrogen sources suggests that this approach could be used to remediate industrial wastewater contaminated with spent dye.

Mikael Eriksson, Erik Sodersten, Zhongtang Yu, Gunnel Dalhammar, William W. Mohn. (Department of Microbiology and Immunology, University of British Columbia, Vancouver, British Columbia V6T 1Z3, Canada. Department of Biotechnology, Royal Institute of Technology, KTH, SE-100 44 Stockholm, Sweden). Degradation of Polycyclic Aromatic Hydrocarbons at Low Temperature under Aerobic and Nitrate-Reducing Conditions in Enrichment Cultures from Northern Soils. *Applied and Environmental Microbiology*, 69(1) (2003), 275-284.

The potential for biodegradation of polycyclic aromatic hydrocarbons (PAHs) at low temperature and under anaerobic conditions is not well understood, but such biodegradation would be very useful for remediation of polluted sites. Biodegradation of a mixture of 11 different PAHs with two to five aromatic rings, each at a concentration of 10 µg/ml, was studied in enrichment cultures inoculated with samples of four northern soils. Under aerobic conditions, low temperature severely limited PAH biodegradation. After 90 days, aerobic cultures at 20°C removed 52 to 88% of the PAHs. The most extensive PAH degradation under aerobic conditions at 7°C, 53% removal, occurred in a culture from creosote-contaminated soil. Low temperature did not substantially limit PAH biodegradation under nitrate-reducing conditions. Under nitrate-reducing conditions, naphthalene, 2-methylnaphthalene, fluorene, and phenanthrene were degraded. The most extensive PAH degradation under nitrate-reducing conditions at 7°C, 39% removal, occurred in a culture from fuel-contaminated Arctic soil. In separate transfer cultures from the

above Arctic soil, incubated anaerobically at 7°C, removal of 2-methylnaphthalene and fluorene was stoichiometrically coupled to nitrate removal. Ribosomal intergenic spacer analysis suggested that enrichment resulted in a few predominant bacterial populations, including members of the genera *Acidovorax*, *Bordetella*, *Pseudomonas*, *Sphingomonas*, and *Variovorax*. Predominant populations from different soils often included phylotypes with nearly identical partial 16S rRNA gene sequences (i.e., same genus) but never included phylotypes with identical ribosomal intergenic spacers (i.e., different species or subspecies). The composition of the enriched communities appeared to be more affected by presence of oxygen, than by temperature or source of the inoculum.

Mishra S, Jyot J. Oilzapper: an oily sludge degrading bacterial consortium for in situ biodegradation of oily sludge. In *Proceedings of the 40th Annual Meet of Association of Microbiologists of India, (2000), 139.*

Huge volume of oily sludge (hazardous waste) is generated during refining of crude oil, the safe disposal of which is a major problem. The use of bacteria in breakdown and digestion of oily sludge is a biological process known as biodegradation. The management of oily sludge through biodegradation is the most eco-friendly and economically viable among the available methods of sludge management. An efficient oily sludge degrading bacterial consortium has been developed. The developed bacterial consortium was immobilized for large-scale application, immobilized bacterial consortium was designated as Oilzapper, could degrade various fractions of oily sludge at faster rate under laboratory and field conditions. In the present study, Oilzapper was used at large scale for biodegradation of oily sludge at an oil refinery. One acre treatment site was taken at an oil refinery for biodegradation of oily sludge. Approximate 300 tonnes oily sludge was spread in treatment site. Oilzapper was applied over the sludge. The oily sludge and Oilzapper was mixed by tilling using a tractor. Based on sludge gradient, treatment site was divided into four blocks. Block A, B and C were treated with Oilzapper while Block D was maintained as untreated control. Block A treated with Oilzapper plus nutrient showed 9.92% contamination of total petroleum carbon (TPH) in soil just after addition of 300 tonne sludge (zero time). It was reduced to 1.63% and 0.97% after 55 days and 120 days respectively indicating 83.5% and 90.92% biodegradation of oily sludge in 55 days and 120 days respectively. In Block B at zero time (just after addition of 300 tonnes oily sludge), 7.61% contamination of TPH (solvent extractable) was recorded. With application of Oilzapper, the TPH in soil reduced from 7.61% to 1.05% and 0.84% after 55 days and 120 days respectively which accounted for 86.2% and 88.9% biodegradation of oily sludge in 55 days and 120 days respectively. Block C was also treated with Oilzapper plus nutrient, which showed 6.45%, 3.11% and 0.25% contamination of TPH in soil at zero time, after 55 days and 120 days respectively which revealed 51.7% and 96% biodegradation of TPH. Block D, which was maintained as untreated control block showed 3.68% contamination of TPH at zero time. The contamination of sludge in Block D reduced from 3.68% to 3.26% after 55 days and 3.06% after 120 days which resulted in biodegradation of sludge only 11.4% and 16.8% in 55 days and 120 days respectively.

N. Perron, U. Welander. (Department of Biotechnology, Center for Chemistry and Chemical Engineering, Lund University, P.O. Box 124, SE-221 00, Lund, Sweden). Degradation of phenol and cresols at low temperatures using a suspended-carrier biofilm process. *Chemosphere, 55(1) (2004), 45-50.*

Degradation of phenol and o-, m- and p-cresol at a concentration of 150 mg l⁻¹ of each compound was studied in a suspended-carrier biofilm process consisting of two aerobic stages. The fungus *Mortierella sarnyensis* Milko dominated the microflora in the first reactor, while bacteria dominated in the second reactor. The process was studied at 4, 7, 11 and 15 °C. The results from the experiments showed the process to be relatively efficient

even at 4 °C. The degradation rate was 33% of that at 15 °C for o-cresol. Both phenol and the cresols were degraded in the first reactor and a new peak appeared in the HPLC-chromatograms indicating the formation of one or more intermediate compounds in the first stage. These compounds were however degraded to below the detection limit in the second reactor. Small new peaks appeared in the chromatograms of the outlet from the second reactor at the maximum loading rates.

Nakamura N., Nakano K., Sugiura N., Matsumura M. A Novel Control Process of Cyanobacterial Bloom using Cyanobacteriolytic Bacteria Immobilized in Floating Biodegradable Plastic Carriers. *Environmental Technology*, 24(12) (2003), 1569-1576.

A process using a floating carrier for immobilization of cyanobacteriolytic bacteria, *B.cereus* N-14, was proposed to realize an effective in situ control of natural floating cyanobacterial blooms. The critical concentrations of the cyanobacteriolytic substance and *B.cereus* N-14 cells required to exhibit cyanobacteriolytic activity were investigated. The results indicated the necessity of cell growth to produce sufficiently high amounts of the cyanobacteriolytic substance to exhibit its activity and also for conditions enabling good contact between high concentrations of the cyanobacteriolytic substance and cyanobacteria. Floating biodegradable plastics made of starch were applied as a carrier material to maintain close contact between the immobilized cyanobacteriolytic bacteria and floating cyanobacteria. The floating starch-carriers could eliminate 99% of floating cyanobacteria in 4 d. Since *B.cereus* N-14 could produce the cyanobacteriolytic substance under the presence of starch and some amino acids, the cyanobacteriolytic activity could be attributed to carbon source fed from starch carrier and amino acids eluted from lysed cyanobacteria. Therefore, the effect of using a floating starch-carrier was confirmed from both view points as a carrier for immobilization and a nutrient source to stimulate cyanobacteriolytic activity. The new concept to apply a floating carrier immobilizing useful microorganisms for intensive treatment of a nuisance floating target was demonstrated.

Naoto Ogawa, Hideo Okamura, Hirofumi Hirai, Tomoaki Nishida. (Department of Forest Resources Science, Faculty of Agriculture, Shizuoka University, Ohya 836, Shizuoka 422-8529, Japan. Faculty of Maritime Sciences, Kobe University, Fukaeminami 5-1-1 Higashinada, Kobe 658-0022, Japan). Degradation of the antifouling compound Irgarol 1051 by manganese peroxidase from the white rot fungus *Phanerochaete chrysosporium*. *Chemosphere*, 55(3) (2004), 487-491.

Irgarol 1051 (2-methylthio-4-tert-butylamino-6-cyclopropylamino-s-triazine), a derivative of s-triazine herbicide, is an antifouling compound used as an alternative to organotins. The compound is highly persistent and is known to be biodegraded only by the white rot fungus, *Phanerochaete chrysosporium*. We used partially purified manganese peroxidase (MnP) prepared from *P. chrysosporium* to evaluate its capacity to degrade Irgarol 1051. MnP degraded Irgarol 1051 to two major products, one identified as M1 (identical to GS26575, 2-methylthio-4-tert-butylamino-6-amino-s-triazine) and the other not identified but with same mass spectrum as M1 and a different ultraviolet spectrum. This report clearly demonstrates that this ligninolytic enzyme is involved in the degradation of Irgarol 1051.

Norbert Scholl. (OXENO Product Safety, Oxeno Olefinchemie GmbH, Paul-Baumann-Strasse 1, P.O. Box 1320, D-45764, Marl, Germany). Ecotoxicity and biodegradation of phthalate monoesters. *Chemosphere*, 53(8) (2003), 921-926.

Little is known about the fate and the effects of phthalic acid monoesters. Various of these monoesters ranging from n-butyl to isononyl monoester have been evaluated in respect to their biodegradation behaviour and their acute aquatic toxicity. All esters are readily biodegradable, achieving degradation rates of 90% and more. The acute toxicity values

strongly depend on the carbon chain length of the alcohol moiety. The short chain specimen have LC/EC₅₀ around and above 100 mg/l, with values levelling off to around 30 mg/l for the isononyl monoester.

Ojumu, T.V., Yu, J. and Solomon, B.O. (Engineering Materials Development Institute, P.M.B 611, Akure, Nigeria. Hawaii Natural Energy Institute, University of Hawaii, Honolulu, HI 96822, USA. Department of Chemical Engineering, Obafemi Awolowo University, Ile-Ife, Nigeria). Production of Polyhydroxyalkanoates, a bacterial biodegradable polymer. African Journal of Biotechnology, 3(1) (2004), 18-24.

There has been considerable interest in the development and production of biodegradable polymer to solve the current problem of pollution caused by the continuous use of synthetic polymer of petroleum origin. Polyhydroxyalkanoates (PHAs) are known to be accumulated as intracellular inclusion in some bacteria. The materials properties exhibited by PHAs, ranging from stiff, brittle to rubber-like makes it a close substitute for the synthetic plastic. The high cost of PHAs production has restricted its applications. The possibility of producing this polymer commercially and at comparable cost has been the main focus in this area.

P. Guiraud, D. Villemain, M. Kadri, O. Bordjiba, R. Steiman. (Groupe pour l'Etude du Devenir des Xénobiotiques dans l'Environnement (GEDEXE), EA 2945 Environnement-Santé, UFR de Pharmacie de Grenoble, Université Joseph Fourier, BP 138, 38243, Meylan cedex, France. Laboratoire d'Etude des Radiopharmaceutiques (LER), INSERM E00-08, UFR. de Médecine de Grenoble, Université Joseph Fourier, 38706, La Tronche cedex, France. Institut des Sciences de la Nature, Université d'Annaba, 23000, Annaba, Algeria). Biodegradation capability of *Absidia fusca* Linnemann towards environmental pollutants. Chemosphere, 52(4) (2003), 663-671.

The purpose of this work was to study the bioremediation capability of *Absidia fusca* Linnemann (Zygomycete) towards different classes of xenobiotics (lignin-derived compounds, chloroaromatic compounds, polycyclic aromatic hydrocarbons) the presence of which in contaminated soils, water and sediments poses a significant risk to the environment and human health. Two strains from different origins were compared. One was from an official collection and grown in non-inducing conditions, while the other was isolated during the course of the survey of fungal flora in a polluted soil from Annaba (Algeria). All data were analyzed and results validated via a statistical treatment. We showed the effect of the factors studied (origin of the strain, xenobiotic) but also the interactions between these factors. The strain of *A. fusca* isolated from a polluted soil was able to efficiently degrade most of the xenobiotics tested, particularly: pentachlorophenol, phenol, catechol, guaiacol and ferulic acid. This property also existed in the other strain but at a very low level.

R. Borja, B. Rincón, F. Raposo, E. Sánchez, A. Martín. (Instituto de la Grasa (CSIC), Avda Padre García Tejero 4, 41012, Sevilla, Spain. Departamento de Ingeniería Química, Facultad de Ciencias, Campus Universitario de Rabanales, Edificio C3, Ctra Madrid-Cádiz, Km 396,14071 Córdoba, Spain). Assessment of kinetic parameters for the mesophilic anaerobic biodegradation of two-phase olive pomace. International Biodeterioration & Biodegradation, 53(2) (2004), 71-78.

A kinetic study of the anaerobic biodegradation of two-phase olive pomace (TPOP) was carried out using a laboratory-scale stirred tank reactor. The reactor was operated at 35°C. The influent contained between 20% and 100% TPOP, representing in terms of total chemical oxygen demand (TCOD) 34.5–187.9 g TCOD l⁻¹. The hydraulic retention times (HRTs) were set between 40.0 and 8.3 d. It was found that the increase of influent

substrate concentration favoured the process failure reducing the pH and increasing the ratio of the total volatile fatty acid (TVFA) to alkalinity. This ratio was found to be proportional to the substrate concentration (S), as follows: $TVFA/alkalinity=0.04(S)$. The kinetic model of Andrews was used to describe the relation between anaerobic biodegradation of TCOD and Volatile Solids (VS) and the formation of methane. The values of the kinetic constants for TCOD removal were determined to be $28 \text{ g TCOD l}^{-1} \text{ d}^{-1}$, 27 g TCOD l^{-1} and $352 \text{ g TCOD l}^{-1}$, respectively, for maximum substrate utilization rates (R_{Smax}), saturation constant (K_S) and inhibition constant (K_i). Process inhibition started at substrate concentrations of around 20 g TCOD l^{-1} . For VS biodegradation the kinetic constant values, R_{Smax} , K_S and K_i , were $45 \text{ g VS l}^{-1} \text{ d}^{-1}$, 37 and 36 g VS l^{-1} , respectively. Inhibition started at VS concentration of around 18 g l^{-1} . The rates of TCOD removal were lower than those observed for VS removal and inhibition of VS removal occurred at a lower concentration compared to that for TCOD. The $Q_{M(max)}$, K_S and K_i constants for methane production were approximately $3.1 \text{ l CH}_4 \text{ l}^{-1} \text{ reactor d}^{-1}$, $8.7 \text{ g TCOD l}^{-1}$ and $272 \text{ g TCOD l}^{-1}$, respectively. Inhibition of methane formation started at a substrate concentration of around 17 g TCOD l^{-1} . TCOD and VS removal rates were higher than the rate of methane formation and these differences increased when the substrate concentration increased. This fact was underlined by the decrease of pH, the increase of TVFA/alkalinity ratio and the reduction of methane production rate.

R. Margesin, D. Labbé, F. Schinner, C. W. Greer, L. G. Whyte. (Institute of Microbiology, University of Innsbruck, A-6020 Innsbruck, Austria. NRC—Biotechnology Research Institute, Montreal, Quebec, Canada H4P 2R2. Department of Natural Resource Sciences, McGill University, Ste. Anne de Bellevue, Quebec, Canada H9X 3V9). Characterization of Hydrocarbon-Degrading Microbial Populations in Contaminated and Pristine Alpine Soils. Applied and Environmental Microbiology, 69(6) (2003), 3085-3092.

Biodegradation of petroleum hydrocarbons in cold environments, including Alpine soils, is a result of indigenous cold-adapted microorganisms able to degrade these contaminants. In the present study, the prevalence of seven genotypes involved in the degradation of n-alkanes (*Pseudomonas putida* GPo1 alkB; *Acinetobacter* spp. alkM; *Rhodococcus* spp. alkB1, and *Rhodococcus* spp. alkB2), aromatic hydrocarbons (*P. putida* xylE), and polycyclic aromatic hydrocarbons (*P. putida* ndoB and *Mycobacterium* sp. strain PYR-1 nidA) was determined in 12 oil-contaminated (428 to 30,644 mg of total petroleum hydrocarbons [TPH]/kg of soil) and 8 pristine Alpine soils from Tyrol (Austria) by PCR hybridization analyses of total soil community DNA, using oligonucleotide primers and DNA probes specific for each genotype. The soils investigated were also analyzed for various physical, chemical, and microbiological parameters, and statistical correlations between all parameters were determined. Genotypes containing genes from gram-negative bacteria (*P. putida* alkB, xylE, and ndoB and *Acinetobacter* alkM) were detected to a significantly higher percentage in the contaminated (50 to 75%) than in the pristine (0 to 12.5%) soils, indicating that these organisms had been enriched in soils following contamination. There was a highly significant positive correlation ($P < 0.001$) between the level of contamination and the number of genotypes containing genes from *P. putida* and *Acinetobacter* sp. but no significant correlation between the TPH content and the number of genotypes containing genes from gram-positive bacteria (*Rhodococcus* alkB1 and alkB2 and *Mycobacterium* nidA). These genotypes were detected at a high frequency in both contaminated (41.7 to 75%) and pristine (37.5 to 50%) soils, indicating that they are already present in substantial numbers before a contamination event. No correlation was found between the prevalence of hydrocarbon-degradative genotypes and biological activities (respiration, fluorescein diacetate hydrolysis, lipase activity) or numbers of culturable hydrocarbon-degrading soil microorganisms; there also was no correlation between the numbers of hydrocarbon

degraders and the contamination level. The measured biological activities showed significant positive correlation with each other, with the organic matter content, and partially with the TPH content and a significant negative correlation with the soil dry-mass content ($P < 0.05$ to 0.001).

René van Herwijnen, Dirk Springael, Pieter Slot, Harrie A. J. Govers, John R. Parsons. (Richard Wasserbauer. (Faculty of Civil Engineering, Czech Technical University in Prague, Thákurova 7, 166 29, Praha 6, Czech Republic). Microbial biodeterioration of electrotechnical insulation materials. International Biodeterioration & Biodegradation, 53(3) (2004), 171-176.

The growth of fungi on insulant surfaces causes change in the electroinsulating properties of the insulant. The paper specifies the mechanisms of deterioration of the surface and internal electric resistances by the micromycetes and the effects of fungi and bacteria on functional characteristics of passive electrical components and transformer oils.

Rincón N.; Chacín E., Marín J., Torrijos M., Moletta R., Fernández N. Anaerobic Biodegradability of Water Separated from Extracted Crude Oil. Environmental Technology, 24(8) (2003), 963-970.

A study of the anaerobic biodegradability of the three categories of water separated from extracted crude oil (Venezuelan oilfields) - light, medium or heavy crude - was carried out at laboratory scale using UASB reactors working at mesophilic conditions. Chemical oxygen demand (COD) removal in a low loaded UASB reactor fed with water separated from extracted light crude was high, with an average 87 % purification efficiency. The remaining COD was made up of the non-biodegradable and the very slowly biodegradable fractions of the organic matter in the water. During a second period, the hydraulic retention time was reduced in stages, thus increasing the loading rate. In the experimental conditions used, COD concentration at the outlet remained below the Venezuelan standard limit for discharge into the environment ($350 \text{ mg COD l}^{-1}$) when the hydraulic retention time (HRT) was above 10 hours and the OLR under $3 \text{ g COD l}^{-1} \text{ d}^{-1}$. For HRT less than 10 hours, or organic loading rate (OLR) greater than $3 \text{ g COD l}^{-1} \text{ d}^{-1}$, COD at the outlet of the reactor rose as a consequence both of increased volatile fatty acids (VFA) concentrations (indicating an overloading of the methanogenic population) and, also, of the increase in the non-VFA COD (indicating a decrease in the acidification efficiency). On the other hand, results with UASB reactors operated at a low loading rate and fed with water separated from extracted medium and heavy crude oil showed that purification efficiency was low, with only 20 % and 37 % COD removal respectively. Continuing the operation of the UASB reactor fed with water separated from medium oil over a prolonged period did not bring any improvement, indicating that no adaptation of the sludge occurred.

S. Hwang and T. J. Cutright. (Department of Civil Engineering, University of Akron, Akron, OH 44325-3905, USA). Biodegradability of aged pyrene and phenanthrene in a natural soil. Chemosphere, 47(9) (2002), 891-899.

A study was conducted to evaluate the biodegradability of pyrene (PYR) and phenanthrene (PHE) aged in a natural soil. Both the single and binary systems were either biostimulated via a nutrient amendment or bioaugmented via an inoculation of the enriched bacteria and nutrients. Aging resulted in higher concentration of both compounds and smaller bacterial activity in the solution-phase. Surprisingly, the total biodegraded extent was greater in the aged soil system than in the freshly spiked system. As anticipated, biostimulation was not appropriate to attain an effective biodegradation in this study, and bioaugmentation achieved a substantial increase the total biodegradation extent. The above findings were attributed to indigenous *Pseudomonas aeruginosa* entering a stationary-phase during the 200-day aging and producing rhamnolipid biosurfactants. In addition, a different sampling

technique (i.e., after vigorous hand-shaking) revealed a 15 times higher microbial population than the normal sampling from the stagnant solution. Therefore, PAH bioavailability in the aged soils can be underestimated when the microbial activity is determined only from the stagnant solution. Furthermore, cometabolism enhanced PYR degradation when PHE was present as a primary substrate.

Sandra Domenek, Pierre Feuilloley, Jean Gratraud, Marie-Hélène Morel, Stéphane Guilbert. (Laboratory of Cereal Technology and Agropolymers, ENSA.M--INRA, 2 place Viala, 34060, Montpellier Cedex 1, France. CEMAGREF, 31 rue J.F. Breton, BP 5095, 34033, Montpellier Cedex 1, France). Biodegradability of wheat gluten based bioplastics. *Chemosphere*, 54(4) (2004), 551-559.

A large variety of wheat gluten based bioplastics, which were plasticized with glycerol, were subjected to biodegradation. The materials covered the total range available for the biochemical control parameter F_i , which expresses the percentage of aggregated proteins. This quantity can be related to the density of covalent crosslinks in the wheat gluten network, which are induced by technological treatments. The biodegradability tests were performed in liquid medium (modified Sturm test) and in farmland soil. All gluten materials were fully degraded after 36 days in aerobic fermentation and within 50 days in farmland soil. No significant differences were observed between the samples. The mineralization half-life time of 3.8 days in the modified Sturm test situated gluten materials among fast degrading polymers. The tests of microbial inhibition experiments revealed no toxic effects of the modified gluten or of its metabolites. Thus, the protein bulk of wheat gluten materials is non-toxic and fully biodegradable, whatever the technological process applied.

Sandra Trott, Shirley F. Nishino, Jalal Hawari, Jim C. Spain. (Air Force Research Laboratory, Tyndall Air Force Base, Florida 32403. Biotechnology Research Institute, National Research Council of Canada, Montreal, Quebec H4P 2R2, Canada). Biodegradation of the Nitramine Explosive CL-20. *Applied and Environmental Microbiology*, 69(3) (2003), 1871-1874.

The cyclic nitramine explosive CL-20 (2,4,6,8,10,12-hexanitro-2,4,6,8,10,12-hexaazaisowurtzitane) was examined in soil microcosms to determine whether it is biodegradable. CL-20 was incubated with a variety of soils. The explosive disappeared in all microcosms except the controls in which microbial activity had been inhibited. CL-20 was degraded most rapidly in garden soil. After 2 days of incubation, about 80% of the initial CL-20 had disappeared. A CL-20-degrading bacterial strain, *Agrobacterium* sp. strain JS71, was isolated from enrichment cultures containing garden soil as an inoculum, succinate as a carbon source, and CL-20 as a nitrogen source. Growth experiments revealed that strain JS71 used 3 mol of nitrogen per mol of CL-20.

Sirpa Metsärinne, Päivi Rantanen, Reijo Aksela, Tuula Tuhkanen. (Department of Environmental Sciences, University of Kuopio, P.O. Box 1627, FIN-70211, Kuopio, Finland. Kemira Oyj, Espoo Research Centre, P.O. Box 44, FIN-02271, Espoo, Finland. Environmental Engineering and Biotechnology, Tampere University of Technology, P.O. Box 541, FIN-33101, Tampere, Finland). Biological and photochemical degradation rates of diethylenetriaminepentaacetic acid (DTPA) in the presence and absence of Fe(III). *Chemosphere*, 55(3) (2004), 379-388.

The environmental fate of ethylenediaminetetraacetic acid (EDTA) has been extensively studied, while much less is known about the environmental behaviour of diethylenetriaminepentaacetic acid (DTPA). In this study, it was confirmed that DTPA is persistent toward biodegradation. The biodegradability of DTPA was investigated in the absence and in the presence of Fe(III) by using CO₂ evolution test and Manometric respirometry test. The CO₂ evolution and oxygen uptake of iron-free (DTPA was added as

free acid) and Fe(III)DTPA were less than in inoculum blank. Possible inhibitor effect was analysed by testing biodegradation of sodium benzoate with and without iron-free or Fe(III)DTPA in the Manometric respirometry test. Only slight inhibition was observed when DTPA was added as free acid. Photodegradation of iron-free DTPA and Fe(III)-DTPA complex was studied by using sunlight and UV radiation at the range 315–400 nm emitted by black light lamps. The results indicate that DTPA added as free acid degrades photochemically in humic lake water. Fe(III)DTPA was shown to be very photolabile in humic lake water in the summer; the photochemical half-life was below one hour. Photodegradation products were identified by the mass spectrometric technique (GC-MS). It was shown that photodegradation of Fe(III)DTPA does not result in total mineralization of the compound. Diethylenetriaminetetraacetic acid, diethylenetriaminetriacetic acid, ethylenediaminetriacetic acid, N,N'- and/or N,N-ethylenediaminediacetic acid, iminodiacetate, ethylenediaminemonoacetic acid and glycine were identified as photodegradation products of Fe(III)DTPA. Based on these observations, we propose a photodegradation pathway for Fe(III)DTPA.

Thomas Borch, Per Ambus, Frank Laturnus, Bo Svensmark, Christian Grøn. (Plant Research Department, Risø National Laboratory, DK-4000, Roskilde, Denmark. Department of Chemistry, University of Copenhagen, DK-2100, Copenhagen Ø, Denmark). Biodegradation of chlorinated solvents in a water unsaturated topsoil. *Chemosphere*, 51(2) (2003), 143-152.

In order to investigate topsoils as potential sinks for chlorinated solvents from the atmosphere, the degradation of trichloromethane (CHCl₃), 1,1,1-trichloroethane (CH₃CCl₃), tetrachloromethane (CCl₄), trichloroethene (C₂HCl₃) and tetrachloroethene (C₂Cl₄) was studied in anoxic laboratory experiments designed to simulate denitrifying conditions in water unsaturated topsoil. Active denitrification was demonstrated by measuring the release of ¹⁵N in N₂ to the headspace from added ¹⁵N labeled nitrate. The degradation of chlorinated aliphatic compounds was followed by measuring their concentrations in the headspace above the soil. The headspace concentrations of all the chlorinated solvents except CH₃CCl₃ were significantly (P < 0.05) lower after 41 days in biologically active batches as compared to sterile batches. For the compounds with significantly decreasing headspace concentrations, the decline was the least for CHCl₃ within the 41 days of incubation. The headspace concentrations of trichloro- and tetrachloroethene decreased more than 50% during the first 20 days with no considerable indication of abiotic transformation. While slow abiotic removal was observed, tetrachloromethane was completely biotransformed after 16 days. Based on the results in this study, we conclude that anaerobic topsoils are potential sinks for these contaminants, and that a natural attenuation potential exists, even in water unsaturated topsoils.

Tomonori Ishigaki, Wataru Sugano, Akane Nakanishi, Masafumi Tateda, Michihiko Ike, Masanori Fujita. (Research Center for Material Cycles and Waste Management, National Institute for Environmental Studies, 16-2, Onogawa, Tsukuba, Ibaraki 305-8506, Japan. Department of Environmental Engineering, Graduate School of Engineering, Osaka University, 2-1, Yamada-oka, Suita, Osaka 565-0871, Japan. Department of Environmental Technology, College of Technology, Toyama Prefectural University, 5180 Kurokawa, Kosugi, Imizu, Toyama, 939-0398, Japan). The degradability of biodegradable plastics in aerobic and anaerobic waste landfill model reactors. *Chemosphere*, 54(3) (2004), 225-233.

Degradabilities of four kinds of commercial biodegradable plastics (BPs), polyhydroxybutyrate and hydroxyvalerate (PHBV) plastic, polycaprolactone plastic (PCL), blend of starch and polyvinyl alcohol (SPVA) plastic and cellulose acetate (CA) plastic were investigated in waste landfill model reactors that were operated as anaerobically and

aerobically. The application of forced aeration to the landfill reactor for supplying aerobic condition could potentially stimulate polymer-degrading microorganisms. However, the individual degradation behavior of BPs under the aerobic condition was completely different. PCL, a chemically synthesized BP, showed film breakage under the both conditions, which may have contributed to a reduction in the waste volume regardless of aerobic or anaerobic conditions. Effective degradation of PHBV plastic was observed in the aerobic condition, though insufficient degradation was observed in the anaerobic condition. But the aeration did not contribute much to accelerate the volume reduction of SPVA plastic and CA plastic. It could be said that the recalcitrant portions of the plastics such as polyvinyl alcohol in SPVA plastic and the highly substituted CA in CA plastic prevented the BP from degradation. These results indicated existence of the great variations in the degradability of BPs in aerobic and anaerobic waste landfills, and suggest that suitable technologies for managing the waste landfill must be combined with utilization of BPs in order to enhance the reduction of waste volume in landfill sites.

V. Matus, M. A. Sánchez, M. Martínez, B. González. (Laboratorio de Microbiología, Departamento de Genética Molecular y Microbiología, and Center for Advanced Studies in Ecology and Biodiversity, Facultad de Ciencias Biológicas, Pontificia Universidad Católica de Chile, Santiago. Departamento de Microbiología, Facultad de Ciencias Biológicas, Universidad de Concepción, Concepción, Chile). Efficient Degradation of 2,4,6-Trichlorophenol Requires a Set of Catabolic Genes Related to *tcp* Genes from *Ralstonia eutropha* JMP134(pJP4). *Applied and Environmental Microbiology*, 69(12) (2003), 7108-7115.

2,4,6-Trichlorophenol (2,4,6-TCP) is a hazardous pollutant. Several aerobic bacteria are known to degrade this compound. One of these, *Ralstonia eutropha* JMP134(pJP4), a well-known, versatile chloroaromatic compound degrader, is able to grow in 2,4,6-TCP by converting it to 2,6-dichlorohydroquinone, 6-chlorohydroxyquinol, 2-chloromaleylacetate, maleylacetate, and β -keto adipate. Three enzyme activities encoded by *tcp* genes, 2,4,6-TCP monooxygenase (*tcpA*), 6-chlorohydroxyquinol 1,2-dioxygenase (*tcpC*), and maleylacetate reductase (*tcpD*), are involved in this catabolic pathway. Here we provide evidence that all these *tcp* genes are clustered in the *R. eutropha* JMP134(pJP4) chromosome, forming the putative catabolic operon *tcpRXABCYD*. We studied the presence of *tcp*-like gene sequences in several other 2,4,6-TCP-degrading bacterial strains and found two types of strains. One type includes strains belonging to the *Ralstonia* genus and possessing a set of *tcp*-like genes, which efficiently degrade 2,4,6-TCP and therefore grow in liquid cultures containing this chlorophenol as a sole carbon source. The other type includes strains belonging to the genera *Pseudomonas*, *Sphingomonas*, or *Sphingopixis*, which do not have *tcp*-like gene sequences and degrade this pollutant less efficiently and which therefore grow only as small colonies on plates with 2,4,6-TCP. Other than strain JMP134, none of the bacterial strains whose genomes have been sequenced possesses a full set of *tcp*-like gene sequences.

Volker Riis, Sabine Kleinstuber, and Wolfgang Babel. Influence of high salinities on the degradation of diesel fuel by bacterial consortia. *Canadian Journal of Microbiology*, 49(11) (2003), 713-721.

Microbial communities from three Argentinean saline soils were extracted and tested for their ability to degrade diesel fuel in liquid culture at salinities between 0% and 25%. In each case, the degradation process was continuously monitored by measuring oxygen consumption. Two communities (CR1 and CR2) showed nearly equal degrees of degradation across a salinity range of 0%–10% (the former degrading about 63% of the diesel fuel and the latter about 70% after 53 and 80 d, respectively). Furthermore, the degree of degradation was not significantly lower in the presence of 17.5% salt (58% and 65% degraded, respectively). A third community (El Zorro) showed a maximum turnover at 5%

salt (79% diesel fuel degraded) and significant degradation (66%) at a salinity of 10%. However, the degree of degradation by this community clearly dropped at 0% and 15% salt. None of the communities were able to degrade diesel fuel in the presence of 25% salt, but the living cell counts showed that components of the microbial population survived the long-term exposure. The surviving portion is obviously sufficient to allow substantial restoration of the original community, as verified by the BIOLOG method. Isolates of the CR1 community were identified as members of the genera *Cellulomonas*, *Bacillus*, *Dietzia*, and *Halomonas*. In light of our investigations, the bioremediation of contaminated saline soils should be quite possible if the salinity of the soil water is lower than 15% or if it is reduced below this limit by the addition of water.

Volker Riis, Wolfgang Babel, Oscar Héctor Pucci. (Department of Environmental Microbiology, UFZ Centre for Environmental Research Leipzig-Halle GmbH, Permoserstrasse 15, D-04318, Leipzig, Germany. C.E.I.M.A., National Patagonian University San Juan Bosco, 9000, Comodoro Rivadavia, Argentina). Influence of heavy metals on the microbial degradation of diesel fuel. *Chemosphere*, 49(6) (2002), 559-568.

The degradation of diesel fuel by a microbial community from a soil polluted by heavy metals (h.m.) in the presence of Cu, Ni, Zn, Pb, Cd, Hg and Cr (as chromate) was investigated. Experiments were conducted with soil slurries and the extracted community in liquid cultivation. The concentrations applied were in the sub-mM and mM range. Whereas the slurries displayed no significant effect, degradation in liquid culture was increasingly inhibited by higher metal concentrations. The course of degradation in suspension was demonstrated by the oxygen consumption. The order of toxicity was found to be: Hg>Cr(VI)>Cu>Cd>Ni>Pb>Zn. The absence of any effect for slurries was due to the non-availability of the metals in the soil, and to precipitation or adsorption to the soil in the case of amendment. The paper also includes results on the availability of h.m. and changes to the community after exposure.

Winnie Dejonghe, Ellen Berteloot,¹ Johan Goris, Nico Boon, Katrien Crul, Siska Maertens, Monica Höfte, Paul De Vos, Willy Verstraete, Eva M. Top. (Laboratory of Microbial Ecology and Technology (LabMET). Laboratory of Microbiology. Laboratory of Phytopathology, Ghent University, B-9000 Ghent, Belgium. Department of Biological Sciences, University of Idaho, Moscow, Idaho 83844-3051). Synergistic Degradation of Linuron by a Bacterial Consortium and Isolation of a Single Linuron-Degrading *Variovorax* Strain. *Applied and Environmental Microbiology*, 69(3) (2003), 1532-1541.

The bacterial community composition of a linuron-degrading enrichment culture and the role of the individual strains in linuron degradation have been determined by a combination of methods, such as denaturing gradient gel electrophoresis of the total 16S rRNA gene pool, isolation and identification of strains, and biodegradation assays. Three strains, *Variovorax* sp. strain WDL1, *Delftia acidovorans* WDL34, and *Pseudomonas* sp. strain WDL5, were isolated directly from the linuron-degrading culture. In addition, subculture of this enrichment culture on potential intermediates in the degradation pathway of linuron (i.e., N,O-dimethylhydroxylamine and 3-chloroaniline) resulted in the isolation of, respectively, *Hyphomicrobium sulfonivorans* WDL6 and *Comamonas testosteroni* WDL7. Of these five strains, only *Variovorax* sp. strain WDL1 was able to use linuron as the sole source of C, N, and energy. WDL1 first converted linuron to 3,4-dichloroaniline (3,4-DCA), which transiently accumulated in the medium but was subsequently degraded. To the best of our knowledge, this is the first report of a strain that degrades linuron further than the aromatic intermediates. Interestingly, the rate of linuron degradation by strain WDL1 was lower than that for the consortium, but was clearly increased when WDL1 was coinoculated with each of

the other four strains. *D. acidovorans* WDL34 and *C. testosteroni* WDL7 were found to be responsible for degradation of the intermediate 3,4-DCA, and *H. sulfonivorans* WDL6 was the only strain able to degrade *N,O*-dimethylhydroxylamine. The role of *Pseudomonas* sp. strain WDL5 needs to be further elucidated. The degradation of linuron can thus be performed by a single isolate, *Variovorax* sp. strain WDL1, but is stimulated by a synergistic interaction with the other strains isolated from the same linuron-degrading culture.

Xiangchun Quan, Hanchang Shi, Jianlong Wang, Yi Qian. (State Key Joint Laboratory of Environment Simulation and Pollution Control, Department of Environmental Science and Engineering, Tsinghua University, Beijing 100084, PR, China). Biodegradation of 2,4-dichlorophenol in sequencing batch reactors augmented with immobilized mixed culture. *Chemosphere*, 50(8) (2003), 1069-1074.

2,4-Dichlorophenol (2,4-DCP) degrading mixed culture was immobilized in polyvinyl alcohol gel beads and supplemented to sequencing batch reactors (SBR) to treat 2,4-DCP containing wastewater. Impacts of bioaugmentation level on the performance of bioaugmented systems were studied. Results show that inoculum size affected the start-up time of the SBR systems. For the non-augmented SBR system, nine days was needed for the system to start-up, whereas it only took six, four, three and two days for the SBRs with 1.9%, 3.7%, 5.6% and 9.3% immobilized culture, respectively. In addition, bioaugmented SBR systems demonstrated stronger capacity to cope with high 2,4-DCP shock loading than the control system. The control SBR failed to treat 2,4-DCP at 166 mg/l in influent, while the SBR with 1.9% inoculation could successfully cope with 2,4-DCP at 166 mg/l, but failed at 250 mg/l, and the SBR with 3.7%, 5.6% and 9.3% immobilized culture could successfully degrade 250 mg/l 2,4-DCP in feed. Furthermore, the contributions to the removal of 2,4-DCP by the introduced and indigenous culture in an augmented SBR system at various operation stages were investigated. It was found that augmented culture played the primary role in degrading 2,4-DCP at the beginning of system start-up, but after one-month operation, both the indigenous and the introduced culture posed strong ability to degrade 2,4-DCP.

Xiaoqi Zhang, Paul L. Bishop. (Department of Civil and Environmental Engineering, University of Massachusetts Lowell, One University Avenue, Lowell, MA 01854, USA. University of Cincinnati, Cincinnati, OH 45221-0018, USA). Biodegradability of biofilm extracellular polymeric substances. *Chemosphere*, 50(1) (2003), 63-69.

This study discovered that biofilm extracellular polymeric substances (EPS) are biodegradable by their own producers and by other microorganisms when they are starved. The study was performed in a comparative fashion to examine the biodegradability of biofilm EPS by the microorganisms from the original biofilm (its own producers) and from activated sludge (other microorganisms). Four distinctive phases were observed during EPS biodegradation. In the first phase, instantaneous concentration increases of carbohydrate and protein in the test solutions were observed when EPS was added; in the second phase, easily biodegradable EPS from the added EPS was quickly utilized; in the third phase, microorganisms began to produce soluble EPS, using the minimally biodegradable EPS left from the previously added EPS; in the fourth phase, cells consumed the newly produced EPS and microbial activity gradually stopped. This study suggests that EPS can be used as a substrate, and that the EPS carbohydrate can be utilized faster than the EPS protein. The EPS utilization rates (including carbohydrate and protein) in the activated sludge suspension were greater than those in the biofilm suspension. It may take microorganisms longer to get acclimated to a new nutrient environment if they are in a starved state.

Young Soo Keum and Qing X. Li. (Department of Molecular Biosciences and Bioengineering, University of Hawaii at Manoa, 1955 East-West Road, Ag Sci 218,

Honolulu, HI 96822, USA). Fungal laccase-catalyzed degradation of hydroxy polychlorinated biphenyls. *Chemosphere*, 56(1) (2004), 23-30.

Hydroxy polychlorinated biphenyls (hydroxy PCBs) are toxic metabolites of PCBs. Their toxicity such as strong endocrine disruption demands effective remediation methods. Laccases from *Trametes versicolor* and *Pleurotus ostreatus* were tested to degrade hydroxy PCBs. Optimum pHs for both enzymes were around 4.0. Laccase from *T. versicolor* degrades hydroxy PCBs more rapidly than that from *P. ostreatus*. The enzymatic activities remained little changes in up to 10% organic solvents, but decreased rapidly in more than 10% acetone, acetonitrile or DMSO. Degradation rate constants decreased with increase of chlorination and no degradation was observed with tetra-, penta- and hexa-chloro hydroxy PCBs in non-mediated reactions. However, the tetra- to hexa-chloro hydroxy PCBs were degraded by laccase from *T. versicolor* in the presence of the mediator 2,2,6,6-tetramethylpiperidine-N-oxy radical. The other mediators, 4-ethyl-2-methoxyphenol, 2,2'-azino-bis(3-ethylbenzthiazoline sulfonic acid) diammonium salt and 1-hydroxybenzotriazole and humic acid, also enhanced degradation of all the hydroxy PCBs except 4-hydroxy-2',3,3',4',5,5'-hexachlorobiphenyl. The results showed that 3-hydroxy biphenyl was more resistant to laccase degradation than 2- or 4-hydroxy analogues. Significant linear-correlations (coefficient of determination, $r^2=0.9097$ and 0.8186 for laccases from *P. ostreatus* and *T. versicolor*, respectively) were found between the ionization potentials and the removal rate constants of hydroxy PCBs.

Zhanpeng Jiang, Hongwei Yang, Lixin Sun, Shaoqi Shi. (Department of Environmental Science and Engineering, Tsinghua University, Beijing 100084, China). Integrated assessment for aerobic biodegradability of organic substances. *Chemosphere*, 48(1) (2002), 133-138.

In this paper, the amount of oxygen consumption, end products and activities of microorganisms are considered as the three factors which affect the biodegradability of organic substances in water. Two integrated assessment methods for biodegradability of organic substances, fuzzy clustering integrated assessment and weighted integrated assessment, have been developed. Simultaneously, the detail steps for assessing a new organic substance biodegradability with these two integrated assessment methods are proposed.

Bioenergy

A. M. Omer, Y. Fadalla. (NCMWE, P.O. BOX 15007, Khartoum 12217, Sudan). Biogas energy technology in Sudan. *Renewable Energy*, 28(3) (2003), 499-507.

Biogas from biomass appears to have potential as an alternative energy in Sudan, which is potentially rich in biomass resources. This is an overview of some salient points and perspectives of biogas technology in Sudan. The current literature is reviewed regarding the ecological, social, cultural and economic impacts of biogas technology. Sudan is blessed with abundant solar, wind, hydro, and biomass resources. Results suggest that biogas technology must be encouraged, promoted, invested, implemented, and demonstrated, but especially for remote rural areas.

Abha Chhabra, S. Palria, V. K. Dadhwal. (Agricultural Resources Group, Remote Sensing Applications Area, Space Applications Centre (ISRO), Ahmedabad 380 015, India. Department of Environmental Studies, M.D.S. University Ajmer 305 009, India). Growing stock-based forest biomass estimate for India. *Biomass and Bioenergy*, 22(3) (2002), 187-194.

The total standing biomass (including above ground and below ground) in Indian forests for the year 1992–93 was estimated using information on state and union-territory field inventory based growing stock volume and the corresponding area under three different crown density classes (very dense forests with crown cover 70 percent and above, dense forest with crown cover 40 percent but <70 percent and open forests with crown cover between 10 and 40 percent) grouped under four major forest categories (hardwood, spruce-fir, pine and bamboo) by Forest Survey of India. The growing stock volume was converted to total biomass using biomass expansion factors as function of growing stock volume density. The average growing stock volume density in Indian forests for the study year 1992–93 was 74.42 m³ ha⁻¹ but it varied amongst states, with a range of 7.1 m³ ha⁻¹ in Punjab to 224.5 m³ ha⁻¹ in Jammu and Kashmir. The total standing biomass (above ground and below ground) was estimated as 8683.7 Mt (Mt=10¹² g). The aboveground and belowground biomass was estimated as 6865.1 and 1818.7 Mt, contributing 79 and 21 percent to the total biomass, respectively. The mean biomass density in Indian forests was estimated as 135.6 tha⁻¹ and amongst the states it varied from 27.4 tha⁻¹ in Punjab to 251.8 tha⁻¹ in Jammu and Kashmir, respectively. The estimates have been compared with previous studies, which had estimated biomass in the range of 4400–8700 Mt for the corresponding period. Our results are an improvement over previous estimates as these incorporate biomass expansion factors which relate wood volume to biomass as a function of growing stock volume density, four forest types and three crown density classes of Indian forests. These improved biomass estimates are crucial to assess the total C pool of forests and further for use as inputs to models to estimate net C flux to atmosphere from Indian forests due to deforestation and landuse changes.

Atul Kumar, Pallav Purohit, Santosh Rana and Tara Chandra Kandpal. (Centre for Energy Studies, Indian Institute of Technology Delhi, Hauz Khas, New Delhi-110016, India). An approach to the estimation of the value of agricultural residues used as biofuels. Biomass and Bioenergy, 22(3) (2002), 195-203.

A simple demand side approach for estimating the monetary value of agricultural residues used as biofuels is proposed. Some of the important issues involved in the use of biomass feedstocks in coal-fired boilers are briefly discussed along with their implications for the maximum acceptable price estimates for the agricultural residues. Results of some typical calculations are analysed along with the estimates obtained on the basis of a supply side approach (based on production cost) developed earlier. The prevailing market prices of some agricultural residues used as feedstocks for briquetting are also indicated. The results obtained can be used as preliminary indicators for identifying niche areas for immediate/short-term utilization of agriculture residues in boilers for process heating and power generation.

BridgwaterAV, Toft A J, and Brammer J G. (Bio-Energy Research Group, Aston University, Birmingham B4 7ET, UK). Power production by biomass fast pyrolysis versus gasification and combustion: a techno-economic comparison. Renewable and Sustainable Energy Reviews, 6(3) (2002), 181-248.

This paper presents an assessment of technical and economic performance of thermal processes to generate electricity from a wood chip feedstock by combustion, gasification and fast pyrolysis. Models were developed to give cost and performance data for integrated system, and pressure gasification, and fast pyrolysis. The models calculated system efficiencies, capital costs, and production costs. The results show that in the long term fast pyrolysis and diesel engine system have great potential to generate electricity at a profit, and at a lower cost than any other biomass to electricity system at small scale.

Christoph Bausch, Matthew Ramsey, Tyrrell Conway. (Department of Botany and Microbiology, University of Oklahoma, Norman, Oklahoma 73019). Transcriptional

Organization and Regulation of the L-Idonic Acid Pathway (GntII System) in Escherichia coli. Journal of Bacteriology, 186(5) (2004), 1388-1397.

The genetic organization of the *idn* genes that encode the pathway for L-idonate catabolism was characterized. The monocistronic *idnK* gene is transcribed divergently from the *idnDOTR* genes, which were shown to form an operon. The 215-bp regulatory region between the *idnK* and *idnD* genes contains promoters in opposite orientation with transcription start sites that mapped to positions -26 and -29 with respect to the start codons. The regulatory region also contains a single putative *IdnR/GntR* binding site centered between the two promoters, a CRP binding site upstream of *idnD*, and an UP element upstream of *idnK*. The genes of the L-idonate pathway were shown to be under catabolite repression control. Analysis of *idnD*- and *idnK*-*lacZ* fusions in a nonpolar *idnD* mutant that is unable to interconvert L-idonate and 5-ketogluconate indicated that either compound could induce the pathway. The L-idonate pathway was first characterized as a subsidiary pathway for D-gluconate catabolism (GntII), which is induced by D-gluconate in a GntI (primary gluconate system) mutant. Here we showed that the *idnK* and *idnD* operons are induced by D-gluconate in a GntI system mutant, presumably by endogenous formation of 5-ketogluconate from D-gluconate. Thus, the regulation of the GntII system is appropriate for this pathway, which is primarily involved in L-idonate catabolism; the GntII system can be induced by D-gluconate under conditions that block the GntI system.

Dogru M, Howarth C R, Akay G, Keskinler B, Malik A A. (Department of Chemical and Process Engineering, University of Newcastle, Newcastle Ne17RU, UK). Gasification of hazelnut shells in a downdrafr gasifier. Renewable Energy, 27(5) (2002), 415-427.

The potential of biomass to reduce GHG (greenhouse gas) is now being recognized widely. Biomass energy is released either by direct combustion or by upgrading into more valuable and useable product such as gas, fuel oil, or higher value products (for utilizations in the chemical industry or for clean power generation). Up till now, gasification work has been concentrated on woody biomass but recently other sources of biomass with large energy production potential have been identified, namely hazelnut shells. Therefore, a pilot scale downdraft gasifier is used to investigate gasification potential of hazelnut shells. A full mass balance is reported including the tar production rate as well as the composition of the produced gas as a function of feed rate. Additionally, the effect of feed rate on the GCV/composition of the product gas and the associated variations of gasifier zone temperature are determined with temperatures recorded throughout the main zones of the gasifier and also at the gasifier outlet and gas cleaning zones. Pressure drops are also measured across the gasifier and gas cleaning system because the produced gas may be used in conjunction with a power production engine when it is important to have low pressure drop in the system. The when it is important to have low pressure drop in the system. The quality of product gas is found to be dependent on the smooth flow of the fuel and the uniformity of the pyrolysis, and so the difficulties encountered during the experiments are detailed. The optimum operation of the gasifier is found to be between 1.44 and 1.47 N m³/kg of air fuel ratios at the values of 4.06 and 4.48 kg/h of wet feed rate which produces the producer gas with a good GCV about 5 MJ/m³ at a volumetric flow of 8-9 N m³/h product gas. It was concluded that hazelnut shells could be easily gasified in a downdraft gasifier to produce good quality gas with minimum polluting by-products.

Francisco Jurado, Antonio Canoa, José Carpiob. (University of Jaén, Department of Electrical Engineering, 23700 EUP Linares (Jaén), Spain. Universidad Nacional de Educación a Distancia, Dept. of Electrical and Computer Engineering, 28040 UNED, Madrid, Spain). Modelling of combined cycle power plants using biomass. Renewable Energy, 28(5) (2003), 743-753.

The olive tree in Spain can generate large quantities of by-product biomass suitable for gasification. Gasification technologies under development would enable these fuels to be used in gas turbines. Biomass conversion to a clean essentially ash-free form, usually by gasification and purification, is necessary to obtain high efficiency. This paper reports results of detailed full-load performance modelling of cogeneration systems based on gasifier/gas turbine technologies.

G. Chen, J. Andries, H. Spliethoff. (Section Thermal Power Engineering, Faculty of Design, Engineering and Production, Delft University of Technology, Mekelweg 2, 2628 CD, Delft, The Netherlands). Biomass conversion into fuel gas using circulating fluidised bed technology: the concept improvement and modelling discussion. *Renewable Energy*, 28(6) (2003), 985-994.

Fuel gas production from biomass using circulating fluidised bed technology is presented in our laboratory. This improved technical concept is aiming at producing high quality gas, in terms of low tar level and particulates carried out in the fuel gas, and overall emissions' reduction associated with fuel gas combustion, as well as stable and reliable operation with the minimum fluctuations in the producer gas volume and composition. Based on this concept, a characteristic theoretical modelling approach involving hydrodynamics, chemical reaction kinetics, and energy balance is accordingly discussed. In addition, very preliminary experimental results from a laboratory-made test rig are also given.

God B. Two phase biomethanation of spent tea leaves for Biogas and manure generation. *Bioresource Technology*, 80 (2001), 153-150.

Anaerobic digestion of spent tea leaves from an instant tea manufacturing factory was two-phase digester. The hydrolysis and acidification phase resulted in the formation of high organic strength liquid called leachate, with a chemical oxygen demand (COD) of 12,880 mg/l, within the retention time of 10 days. The leachate was tested in a batch methanogenesis reactor for biogas production. An average biogas yield of 0.48 m³/kg of COD destroyed was obtained with an average COD reduction of 93%.

H. Rezai, F. M. Yusoff, A. Kawamura, A. Arshad, B. H. R. Othman. Zooplankton biomass in the Straits of Malacca. *Indian Journal of Marine Sciences*, 32(3) (2003), 222-225.

The distribution patterns of zooplankton biomass were studied using samples collected in vertical hauls during four oceanographic cruises in the Straits of Malacca between November 1998 and August 2000 with 140 µm-mesh and 45-cm diameter NORPAC net. The average zooplankton biomass during Cruise III (post-SW monsoon) and IV (SW monsoon) was almost twice that of Cruise I (NE monsoon) with maximum zooplankton biomass occurring during the Cruise IV. Biomass was generally higher in waters closer to the near-coastal areas than in the neritic areas. Higher zooplankton biomass values occurred in the central part of the Straits compared to other areas, although spatial and temporal variations of biomass were not significant. Run off of major rivers and the extensive mangrove forests in the coastal areas might have influenced higher biomass in the central part of the Straits.

J. W. Goodrum. (Department of Biological and Agricultural Engineering, University of Georgia, Athens, GA 30602, USA). Volatility and boiling points of biodiesel from vegetable oils and tallow. *Biomass and Bioenergy*, 22(3) (2002), 205-211.

Quality control of fuel-related properties of Biodiesel, such as volatility, is needed to obtain consistent engine performance by fuel users. The vapor pressures and boiling points of selected methyl esters and vegetable oils are proposed as quality control metrics for Biodiesel. This type of data was obtained by a rapid new method using thermogravimetric analysis (TGA). One atmosphere boiling points (bps) and temperature-dependent vapor

pressures from 1 atm down to 5.332 kPa (40 mmHg) were measured for methyl and ethyl esters of rapeseed oil, canola oil, soybean oil, and tallow. Boiling points (1 atm) ranged from 340°C to 375°C. Methyl and ethyl esters of a given oil differ by ca. 5°C in bps. These results are discussed in terms of the fatty acid composition of the esters and oils. Calibrations showed that the TGA/laser orifice capsule method gave accuracy of $\pm 5\%$.

Jan Erik Mattsson, Pieter D. Kofman. (Department of Agricultural Engineering, Swedish University of Agricultural Sciences, SE-230 53 Alnarp, Sweden. Danish Forest and Landscape Research Institute, Kvak Moellevej 31, DK-7100 Vejle, Denmark). Method and apparatus for measuring the tendency of solid biofuels to bridge over openings. Biomass and Bioenergy, 22(3) (2002), 179-185.

The paper describes the development of a method to measure the relative bridging properties of solid biofuels over openings. The system consists of a silo, with a bottom made of rubber mats, which can be rolled on a drum, so that a gap is created. Over the gap, a "bridge" of fuel particles is formed. When the gap is wide enough, the bridge collapses and the distance between the drums is measured. This number is a measure of the relative bridging property of that specific fuel. General results are given for a variety of solid biofuels. The bridging property of solid biofuels is mainly controlled by the shape and the size of the particles. If the fuel contains many hooked or long and thin particles, the bridging tendency is high. The influence of the moisture content is not as pronounced, but exists. In further articles, specific results will be discussed.

Meshram J R. (Director, Ministry of Non-conventional Energy Sources, Block 14, CGO Complex, Lodi Road New Delhi- 110003,India). Biomass resource assessment programme and prospects of biomass as an energy source in India. Ireda News, 13(4) (2002), 21-29.

Biomass is a very important renewable energy resource in India. More than 19 500 MW of power can be produced from surplus biomass if adequate price is paid to biomass producers/users. This paper discusses the potential for surplus biomass production and also how much of it can be used to produce power in India.

P. Sudhaa, H. I. Somashekharb, Sandhya Raoc, N. H. Ravindranath. (Centre for Ecological Sciences, Indian Institute of Science, Bangalore, India. Centre for Application of Science and Technology for Rural Areas, Indian Institute of Science, Bangalore, India. Department of Civil Engineering, Indian Institute of Technology, New Delhi, India). Sustainable biomass production for energy in India. Biomass and Bioenergy, 25(5) (2003), 501-515.

The availability of land for biomass production, the various biomass production options, biomass productivity rates, financial viability, investment required to produce biomass for energy and the barriers to biomass production are analysed. The scenarios considered for estimating the biomass potential are incremental biomass demand, sustainable biomass demand and the full biomass demand. Under these scenarios, two situations namely no increase in cropland by 2010 and increase in cropland by 10% over 1995 area have been considered. The land available for biomass production ranges from 9.6 to 36.5 Mha under the different scenarios. Annually 62–310 Mt of wood could be generated from the surplus land, after meeting all the requirements of biomass, such as domestic fuelwood, industrial wood and sawnwood, with an investment of Rs168–780 billion. An electricity generation potential of 62–310 TWh annually is estimated. The key barriers to produce biomass sustainably for energy are lack of commercial demand for wood for energy, lack of financial incentives, low productivity of plantations, land tenurial barriers and lack of institutions to integrate biomass production for energy and bioenergy utilities.

R. Sunil Kumar. Biomass, horizontal zonation and vertical stratification of polychaete fauna in the littoral sediment of Cochin estuarine mangrove habitat, south west coast of India. Indian Journal of Marine Sciences, 31(2) (2002), 100-107.

Biomass distribution, horizontal zonation, relative dominance and vertical distribution of polychaetes were studied. Highest biomass was recorded in the mid tidal region in both study areas. The monthly values varied from 4.43 to 128.28 g.m⁻² at st 1 and 2.57 to 67.31 g.m⁻² at st 2. Multiple regression analysis between biomass and environmental parameters indicate that they could not individually or in combination bring about the spatial and temporal variability in biomass distribution. Moreover, at st 1 edaphic factors appear to be responsible for partial variation in biomass indicating comparatively high F ratio for variance analysis than station 2. ANOVA of species diversity indices values ($P < 0.5$) between the three tidal regions showed a clear horizontal zonation of polychaetes, especially at st 1. A substantial difference in percentage composition of fauna, up to 15 cm depth of mangrove soil, was found in all the three tidal zones studied. This variability in the community structure in the top (0-5 cm) and deeper mangrove sediment (10-15 cm) is pertained to a variety of characteristic features of both upper and deeper sediments. High numerical abundance and coexistence of certain euryhaline species showed significant similarity index (>70%) among polychaete fauna. This similarity and affinity of fauna for a long period evidently indicate the habitat stability that is pertained to the existence of species diversity and abundance. This is related to the prolonged food resource input and profound standing capacity in the littoral mangrove soil, which virtually render in building up a stable community structure of polychaetes. As a result of this, selection of habitat by polychaetes, its survival and subsequent long-term biomass production were occurred. The strong similarity of polychaete fauna between months and biomass productivity can be considered for deriving the productive characteristic of the mangrove habitat, and also for assessing demersal fishery potential.

Rajeshwari V, Kusum Lata, Pant D C, Kishore V V N. A novel process using enhanced enhanced acidification & a USAB reactor for biomethanation of vegetable market waste. Indian Journal of Marine Sciences, 2001, 292-300.

The processing of solid vegetable market waste was attempted in B two-stage digester. Initially for the- hydrolysis and acidification of waste, enhanced extraction process was carried out by continuous sprinkling of water over the bed of waste in a mild steel 100 l solid bed digester. The leachate obtained after completion of acidification phase was further treated in 24.61 UASB reactor for biogas production. The effect of waste-liquid ratio, dilution and sprinkling rate on the digestion of waste were studied. A high strength leachate, with a COD of 25,298 mg l⁻¹ and total volatile fatty acids concentration of 11.3 g l⁻¹ was obtained as a result of hydrolysis and acidification in a very short retention time of 40 h. The COD reduction of 94% was achieved during UASB reactor operation at organic loading rate of 19,6 kg COD m⁻² day⁻¹ a minimum hydraulic retention time of 16 h was achieved.

S. Dapía, V. Santos and J. C. Parajó. (Department of Chemical Engineering, University of Vigo (Campus Ourense), Polytechnical Building, Campus Universitario, As Lagoas, 32004 Ourense, Spain). Study of formic acid as an agent for biomass fractionation. Biomass and Bioenergy, 22(3) (2002), 213-221.

Beech, hardwood samples were treated in 80% formic acid solutions under a variety of experimental organosolv pulping conditions. The effects of the main operational variables on the composition and yield of cellulose pulps obtained in treatments have been assessed. The SCAN viscosity of cellulose pulps was measured in order to determine the damage of the cellulose produced during the treatments, and additional data on the composition of liquors from treatments were determined to assess the type and amount of valuable byproducts.

S. Nonhebel. (IVEM, Center for Energy and Environmental Studies, University of Groningen, Nijenborgh 4, 9747 AG Groningen, Netherlands). Energy yields in intensive and extensive biomass production systems. Biomass and Bioenergy, 22(3) (2002), 159-167.

As for agricultural crops, biomass crops can be grown in intensive production systems (external inputs such as pesticides and artificial fertilisers) or extensive systems with few external inputs. The choice between an intensive or extensive production system has consequences for yields. A method is presented to estimate biomass yields in intensive and/or extensive production systems. This method is applied to a poplar coppice production system. Results of the method are used to evaluate several intensive and extensive production systems with respect to bioenergy yield and fossil fuel use efficiency. The energy yield (GJ/ha) of the intensive systems was highest, while the extensive systems show the better fossil fuel use efficiency (GJ output/GJ fossil energy input).

Smith K R, Uma R, Zhang J, Kishore V, V N, Joshi V. Greenhouse implications of household fuels. An analysis for India Annual review of energy and environment. 19 (2000), 741-765.

It is commonly assumed that biomass fuel cycles based on renewable harvesting of wood or agricultural wastes are greenhouse-gas (GHG) neutral because the combusted carbon in the form of CO₂ is soon taken up by regrowing vegetation. Thus, the two fifths or more of the world's households relying on such fuels are generally not thought to play a significant role in GHG emissions, except where the wood or other biomass they use is not harvested renewably. This review examines this assumption using an emissions database of CO₂, CO, CH₄, NMHC, N₂O, and total suspended particulate emissions from a range of household stoves in common use in India using six biomass stoves are thermally inefficient and divert substantial fuel carbon to products of incomplete combustion, their global warming commitment (GWC) per meal is high. Depending on time horizons and which GHGs are measured, the GWC of a meal cooked on a biomass stove can actually exceed that of the fossil fuels, even if based on renewably harvested fuel. Biogas, being based on a renewable fuel and, because it is a gas, being combusted with high efficiency in simple devices, has by far the lowest GWC emitted at the stove per meal and is indicative of the advantage that upgraded fuels made from biomass have in moving toward sustainable development goals. There are a number of policy implications of this work, including revelation of a range of win-win opportunities for international investment in rural energy development that would achieve cost-effective GHG reduction as well as substantial local benefits.

Srinivas S N. Biomass consumption in unorganized enterprises in India. Biomass Users Network India, 3(3), 2-4.

The unorganized enterprises form a very important sector in India that contribute considerably to economy and exports. A large number of unorganized enterprises use fuels for various processes in their units. The fuel used in these enterprises are, coal, coal gas, coke, firewood, charcoal L P Gas, diesel oil, furnace oil, other fuel oils; electricity, water stream, solar energy, animal power, and others. The unorganized sector has very large number of energy consuming enterprises and the energy consumption has an increasing trend (expenditure on energy inputs is more than proportionate to the increase in expenditure on total inputs). Maximum number of units depend on biomass fuels to meet their energy requirements. The inconsistencies in presentation of data in different survey reports pose hurdles in understanding, the sector better specifically the energy issues. The scantiness in the data is also evident from the gross underestimation of units and perhaps the total consumption of fuels.

Surinder Katyal, Kelly Thambimuthu, Marjorie Valix. (Department of Chemical Engineering, The University of Sydney, NSW, Australia 2006. Present Address: Group Leader & Senior Research Scientist, Fossil Fuels & Climate Change, CANMET Energy Technology Center, Natural Resources Canada, Ottawa, Canada). Carbonisation of bagasse in a fixed bed reactor: influence of process variables on char yield and characteristics. *Renewable Energy*, 28(5) (2003), 713-725.

Carbonisation experiments on samples of sugar cane bagasse were conducted in a static fixed bed reactor to determine the effect of process variables such as temperature, heating rate, inert sweep gas flow rate and particle size on the yield and composition of solid product char. Experiments were performed to the final temperatures of 250–700°C with heating rates from 5 to 30°C/min with nitrogen sweep gas flow rate of 350 cc/min. Additional tests were aimed at studying the effect of different flow rates of nitrogen sweep gas from 0 to 700 cc/min during carbonization and different particle size fractions of bagasse. The results showed that as the carbonisation temperature was increased, the yield of char decreased. The reduction in yield was rapid up to a final temperature of 500°C and was slower thereafter. The yield of char was relatively insensitive to the changes in heating rate and particle size. Increasing the sweep gas flow rate to 350 cc/min reduced the yield of char. It appears the presence of inert sweep gas reduced secondary reactions which promoted char formation. The proximate analysis of the char suggests that fixed carbon and ash content increased with temperature. The char obtained at temperatures higher than 500°C have high carbon content and is suitable as renewable fuel and for other applications. The carbonization of bagasse has the potential to produce environmental friendly fuels and can assist in reducing deforestation for the production of charcoal.

Bioengineering

Abraham F G Stevenson. (Radiation Toxicology Unit, Institute for Experimental Toxicology, Centre for Environmental Sciences, University of Kiel, Brunswiker Str. 10, D-24105 Kiel, Germany). Tissue engineering: In vitro embryonal nidation in a murine endometrial construct. *Indian Journal of Experimental Biology*, 41 (2003), 563-569.

The epithelial and mesothelial cellular components of organs can be obtained as dissociated cells using adequate procedures of enzymatic digestion followed by pycnotic separation on density gradients. Using a specially developed procedure for tissue dissociation, the epithelial and connective tissue components of endometria from pseudopregnant mice were grown in culture using a combination of three dimensional culture of connective tissue components in collagen gel, with the superimposition of epithelial components in liquid medium on the surface of the gells. After a few days of growth, when the cultures became dense, murine blastocysts obtained on postcoital day 4.5 by fallopian flushing of hormonally primed and mated mice, were transferred onto the imitated endometria. The blastocysts hatched and grew on the endometrial epithelium as spherical coherent conglomerates of cells quite different from hatched blastocysts grown on the surface of a petri dish, in which the presumptive trophoblasts spread around the central mass. Light and electronmicroscopy of resin embedded sections (2 days after nidation on the simulated endometria) revealed that at least two populations of cell types were recognisable as layers. This is interpreted as an early sign of morphogenesis and the first visible steps of differentiation. The presence of mitotic figures indicates viability and continuing growth. Electronmicroscopy of cell types grown under conditions simulating in vivo tissue architectonics showed overtly less cytopathology and better cell function. Simulated endometria may, therefore, serve as an attractive model for studying early mammalian embryogenesis and the effects of toxic agents.

Bhattacharya D, Mishra S, Sarma P M. Molecular typing of hydrocarbon degrading bacterial strains. In Proceedings of Bio-horizon. 3rd National Symposium on Biochemical engineering and Biotechnology, (2001), 155.

Oily sludge/crude oil (contamination is a major environmental concern since many of the constituent hydrocarbons are toxic. Besides this incidence of crude oil spill is increasing despite the best effort of petro-chemical industry. Natural diversity of soil microorganisms is a useful source for detoxification of hazardous waste contamination sites. Understanding of the microbial diversity in such an ecosystem will lead to formulation of better strategies of bioremediation. The present study was taken up to estimate the diversity of hydrocarbon degrading organisms. Molecular typing of hydrocarbon degrading bacterial strains was performed to study the genetic diversity among the isolated bacterial strains.

Daljit S. Arora, Paramjit K. Gill. (Microbial Technology Laboratory, Department of Microbiology, Guru Nanak Dev University, Amritsar 143005, India). Comparison of two assay procedures for lignin peroxidase. Enzyme and Microbial Technology, 28(7-8) (2001), 602-605.

The most widely accepted assay for detecting lignin peroxidase, based on the oxidation of veratryl alcohol to veratraldehyde, suffers from some drawbacks. At 310 nm, the wavelength at which the assay is performed, some other materials like lignins, quinonic compounds and aromatics also exhibit strong absorbance thus interfering with the estimation when present in the media. The present study reports the lignin peroxidase production by some white rot fungi under different nutritional conditions. The veratryl alcohol oxidation assay procedure for lignin peroxidase has been compared with another method based on the oxidation of the dye azure B involving absorbance measurements in the visible range. The latter method proved to be much more advantageous over the veratryl alcohol oxidation method, in media supplemented with malt extract, lignin preparations and agricultural residues. The enzyme production by veratryl alcohol assay could be detected only in mineral salts broth. By the azure B assay the enzyme activity was detected in all the media tested. The supplements gave varied response in different media. Veratryl alcohol enhanced the enzyme production in malt extract broth and mineral salts malt extract broth. Among the lignin preparations Indulin AT increased the lignin peroxidase titres from 2 to 20 fold in different fungi. Similarly, wheat straw supplemented in mineral salts broth and malt extract broth, separately, strongly stimulated the lignin peroxidase production. The above studies revealed that azure B assay may act as a substitute or equivalent method.

Daljit Singh Arora, Paramjit Kaur Gill. (Microbial Technology Laboratory, Department of Microbiology, Guru Nanak Dev University, Amritsar, 143005 Punjab, India). Effects of various media and supplements on laccase production by some white rot fungi. Bioresource Technology, 77(1) (2001), 89-91.

White rot fungi produce three main extracellular enzymes involved in ligninolysis; laccase, lignin peroxidase and manganese peroxidase. Though all white rot fungi do not produce all three enzymes, laccase occupies an important place in ligninolysis. The present paper reports its production by some white rot fungi; *Daedalea flavida*, *Phlebia brevispora*, *Phlebia radiata* and *Polyporus sanguineus* under different nutritional conditions. Of the various basal media tested, mineral salts malt extract broth proved to be the best medium for laccase production. Sugarcane bagasse proved to be the best laccase inducer among the various supplements added to different media.

P. Gill, D. Arora. (Department of Microbiology, Guru Nanak Dev University, Amritsar 143 005, Punjab, India). Effect of culture conditions on manganese

peroxidase production and activity by some white rot fungi. *Journal of Industrial Microbiology and Biotechnology*, 30(1) (2003), 28 – 33.

The ligninolytic system of white rot fungi is primarily composed of lignin peroxidase, manganese peroxidase (MnP) and laccase. The present work was carried out to determine the best culture conditions for production of MnP and its activity in the relatively little-explored cultures of *Dichomitus squalens*, *Irpex flavus* and *Polyporus sanguineus*, as compared with conditions for *Phanerochaete chrysosporium* and *Coriolus versicolor*. Studies on enzyme production under different nutritional conditions revealed veratryl alcohol, guaiacol, Reax 80 and Polyfon H to be excellent MnP inducers.

Biofertilizer

A.K. Mukherjee, S.R.Mandal, UDITA Mandal AND B.C. Patra. (Department of agronomy, Bidlnin Chandra Krishi Viswavidyalaya, Mohanpur 741252, India). Effect of Biofertilizer Application on Forage Yield and Quality of Maize Crops. *Environment and Ecology*, 22(1) (2004), 211-214.

A Field trial on neutral sandy loam entisol was conducted of study the influence of *Azotobacter* and *Azospirillum* in combination of different doses of fertilizer-N on maize cv Vijoy composite. Though *Azotobacter* increases significantly both mean green (5%) and (15%) forage yields, *Azospirillum* reduced the yields significantly (15 and 17% respectively). With respect to mean CP, DCP and NDF per cent, both *Azotobacter* (9.88, 2.66, 50.0) and *Azospirillum* (9.98, 3.66, 50.8) inoculation proved superior over control (7.90, 2.19, 45.3). CP DCP and NDF yields are also affected in the similar way. These qualities (both in per cent and yields) also increased with the rate of fertilizer applications (0–120 kg N/hectare) but application of fertilizer N more than 80 kg/hectare was not found much useful and suppressed the beneficial effect of bacteria.

Biomarkers

K. L. Londry, L. L. Jahnke, D. J. Des Marais. (Department of Microbiology, University of Manitoba, Winnipeg, Manitoba, Canada, Exobiology Branch, NASA Ames Research Center, Moffett Field, California). Stable Carbon Isotope Ratios of Lipid Biomarkers of Sulfate-Reducing Bacteria. *Applied and Environmental Microbiology*, 70(2) (2004), 745-751.

We examined the potential use of natural-abundance stable carbon isotope ratios of lipids for determining substrate usage by sulfate-reducing bacteria (SRB). Four SRB were grown under autotrophic, mixotrophic, or heterotrophic growth conditions, and the $\delta^{13}\text{C}$ values of their individual fatty acids (FA) were determined. The FA were usually ^{13}C depleted in relation to biomass, with $\Delta\delta^{13}\text{C}(\text{FA} - \text{biomass})$ of -4 to -17‰; the greatest depletion occurred during heterotrophic growth. The exception was *Desulfotomaculum acetoxidans*, for which substrate limitation resulted in biomass and FA becoming isotopically heavier than the acetate substrate. The $\delta^{13}\text{C}$ values of FA in *Desulfotomaculum acetoxidans* varied with the position of the double bond in the monounsaturated C16 and C18 FA, with FA becoming progressively more ^{13}C depleted as the double bond approached the methyl end. Mixotrophic growth of *Desulfovibrio desulfuricans* resulted in little depletion of the i17:1 biomarker relative to biomass or acetate, whereas growth with lactate resulted in a higher proportion of i17:1 with a greater depletion in ^{13}C . The relative abundances of 10Me16:0 in *Desulfobacter hydrogenophilus* and *Desulfobacterium autotrophicum* were not affected by growth conditions, yet the $\Delta\delta^{13}\text{C}(\text{FA} - \text{substrate})$ values of 10Me16:0 were considerably

greater during autotrophic growth. These experiments indicate that FA $\delta^{13}\text{C}$ values can be useful for interpreting carbon utilization by SRB in natural environments.

L.J.R. Foster, A. Saufi, P.J. Holden. (Department of Biotechnology, The University of New South Wales, UNSW-Sydney NSW 2052, Australia. Australian Nuclear Science & Technology Organisation (ANSTO), Lucas Heights, NSW 2258, Australia). Environmental concentrations of polyhydroxyalkanoates and their potential as bioindicators of pollution. *Biotechnology Letters*, 23(11) (2001), 893-898.

A quick and inexpensive protocol based on gas chromatography was used to identify and measure environmental concentrations of microbial polyhydroxyalkanoates, PHAs. Samples taken from apparently unpolluted sites characterised by agricultural land or native vegetation possessed concentrations ranging from 0.12 to 0.40 mg PHA per g sample. In contrast, environments impacted by anthropogenic activity displayed concentrations 14 to 40 times higher. The results support the suggestion that PHAs could be used as pollution bioindicators in preliminary assessments of environmental health.

Maurizio Aceto, Ornella Abollino, Raffaele Conca, Mery Malandrino, Edoardo Mentasti, Corrado Sarzanini. (Department of Sciences and Advanced Technologies, University of Eastern Piedmont, Spalto Marengo, 33-15100, Alessandria, Italy. Department of Analytical Chemistry, University of Turin, via Giuria, 5-10125, Turin, Italy). The use of mosses as environmental metal pollution indicators. *Chemosphere*, 50(3) (2003), 333-342.

The possibility of using mosses as environmental indicators of metal pollution has been investigated. Mosses of the species *Bryum argenteum* were collected from different parts of Piedmont (Italy), ranging from highly polluted areas to nearly uncontaminated mountain areas. Periodical samplings were planned in every site on a monthly base, in order to check variations of metal uptake throughout one year; correlations with pluviometric and thermal patterns were investigated for all sampling stations. On every moss sample 20 elements, ranging from major (K, P, Al, Ca, Fe and Mg) to minor (Mn, Na, Ti and Zn) and trace (As, Ba, Cd, Co, Cr, Cu, Li, Ni, Pb and Sr), were quantitatively determined by inductively coupled plasma-atomic emission spectrometry or graphite furnace-atomic absorption spectrometry, depending on the needed sensitivity. Statistical analyses, carried out with principal component analysis and cluster analysis methods, revealed that a good correlation exists between metal content in mosses and pollution degree in the areas sampled.

S. Lecoeur, B. Videmann, Ph. Berny. (UMR INRA-DGER Métabolisme et Toxicologie Comparés des Xénobiotiques, Ecole Nationale Vétérinaire de Lyon, 1, av. Bourgelat, BP 83, F-69280, Marcy l'Etoile, France). Evaluation of metallothionein as a biomarker of single and combined Cd/Cu exposure in *Dreissena polymorpha*. *Environmental Research*, 94(2) (2004), 184-191.

The effects of metal mixture (Cd+Cu) versus single-metal exposure on total MT response and bioaccumulation were investigated in the freshwater bivalve *Dreissena polymorpha*. A two-month exposure period, including two levels of contamination, was chosen for each of the two metals: 5, 10 $\mu\text{g/L}$ for Cu, and 2, 20 $\mu\text{g/L}$ for Cd, with mixtures of, respectively, 5 $\mu\text{g/L}$ Cu+2 $\mu\text{g/L}$ Cd, 5 $\mu\text{g/L}$ Cu+20 $\mu\text{g/L}$ Cd, 10 $\mu\text{g/L}$ Cu+2 $\mu\text{g/L}$ Cd, and 10 $\mu\text{g/L}$ Cu+20 $\mu\text{g/L}$ Cd. Total MT contents were assessed by an Ag-saturation method, and metals contents were determined by atomic absorption spectrometry. Results at the whole-organism level showed a significant and early increase of total MT biosynthesis after exposure to Cd. This increase was significantly correlated with Cd bioaccumulation. By contrast, Cu did not modify total MT response, and mussels limited Cu bioaccumulation. The mixture either did not influence or only weakly influenced metal accumulation and MT response to Cu and Cd

after long-term exposure. Our results suggest that the form of MT existing in *D. polymorpha* was not Cu-inducible. This could limit the use of MT in *D. polymorpha* as a biomarker of heavy metal pollution in freshwater ecosystems.

W. J. Reynolds, S. W. Feist, G. J. Jones, B. P. Lyons, D. A. Sheahan, G. D. Stentiford. (The Centre for Environment, Fisheries and Aquaculture Sciences, Remembrance Avenue, Burnham-on-Crouch, Essex CM0 8HA, UK. CEFAS, Barrack Road, The Nothe, Weymouth, Dorset DT4 8UB, UK. CEFAS, Pakefield Road, Lowestoft, Suffolk NR33 0HT, UK). Comparison of biomarker and pathological responses in flounder (*Platichthys flesus* L.) induced by ingested polycyclic aromatic hydrocarbon (PAH) contamination. *Chemosphere*, 52(7) (2003), 1135-1145.

This study investigated the response of biomarker measurements and histopathological indicators of polycyclic aromatic hydrocarbon (PAH) exposure in the flounder (*Platichthys flesus* L.). Flounder were fed food spiked with a mixture of four PAHs at an environmentally relevant range of concentrations for either one or six months. Ethoxyresorufin-O-deethylase (EROD) activity was elevated following 1 month exposure to PAH concentrations up to 50 mg kg⁻¹ in food. Bile metabolite concentrations were found to increase with PAH concentration, up to 500 mg kg⁻¹ PAH. By comparison, no DNA adducts were detected and there were no significant histopathological changes observed. After 6 months exposure, EROD levels were not elevated but bile metabolites showed a similar dose dependent relationship as in the 1 month experiment, while DNA adducts were only detected in the highest PAH exposure groups. No significant histopathological changes were observed. The results are discussed with respect to the implications for the use of these methods in environmental monitoring studies.

Biopesticide

A. Jebanesan, M. R. Venkatachalam, G. Jagadeesan. (Department of Zoology, Annamalai University, Annamalinagar-608002, Tamil Nadu, India). Toxicity evaluation of certain neem-based bio-insecticides against the larval mosquitoes of *Culex quinquefasciatus* (say). *Pestology*, 27(5) (2003), 27-29.

Commercially available neem-based bio-insecticides like Nimbex, Neem Gold, Nimbecidine and Neem Shakthi were selected to study the acute toxicity for larval age groups of *Culex quinquefasciatus*. These bio-insecticides can be placed in the decreasing order of hierarchy for toxicity to different larval age groups of *Culex quinquefasciatus* as Nimbex > Nimbecidine > Neem Gold > Neem Shakthi. Among the different larval age groups of *Culex quinquefasciatus* tested, the lethal response of the 1st instar larvae was more when compared to that of other age groups.

Aarti Mahajan, Suvasish Das. (Ph.D Division of Agricultural Chemical, Indian Agricultural Research Institute, New Delhi-110012. Ph.D School of Environment Management, Guru Gobind Singh Indraprastha University, Delhi-11006). Plants and Microbes - Potential Source of Pesticide for Future Use. *Pesticides Information*, 28(4) (2003), 33-34.

Development of insecticide resistance has become well known among most organochlorine, organophosphorus, carbamates, formamidines and pyrethroid insecticides. This is solely attributed to extensive and indiscriminate use of these chemicals and others restricted in use or prohibited due to potential environmental hazards. In the prevailing scenario, it becomes imperative to direct the presentday research toward synthesis of new molecules, which satisfy well the various prerequisites such as their safety and sustainability aspects. The problem of insecticide resistance aroused due to disturbed ecological balance and

expression of dormant genetic trait or existing polymorphism among target pests. Answer to such problem lies in the nature, which is endowed with enormous treasure in form of biodiversity. Secondary metabolites produced by both plants and microbes in situ which have been found to show antagonistic effect against certain insect pests has been documented for quite sometime and are continuously explored. It is estimated that to date only 10 percent of plant species in the world have been examined chemically. Therefore, a challenging task to discover newer class of pesticides and new mode of action from natural plant source lies beforehand. Several such molecules active against pests find mention in this discussion. Advantages with such molecules are many due to diversity and novelty of chemical structure produced by living organisms and potential specificity of biological actions. For instance, the risk of bioaccumulation of these molecules in groundwater residues is greatly reduced apart from the advantage of overcoming the resistance development. There is greatly reduced likelihood of harmful bioaccumulation on soil and ground water residues in addition to resistance development. Potential of Plants as Source of Pesticides Botanical pesticides can play an important role in controlling pests. Screening of plant preparations, followed by bioassay-guided fractionation, leading to isolation and identification of pure active plant constituents, is considered as one of the most successful methodologies for the discovery of new products.

D. N. Mishra, K. Kumar. (SVBPUA & T, Research Station, Nagina, Bijnor 246762, India). Evaluation of Neem Based Formulations and Neem Seed Kernel Extract Against Insect Pests of Sesamum. Environment and Ecology, 22(1) (2004), 80-84.

The effect of neem based formulations on the population of insect pests of sesamum were studied during 2000–2002. Among various treatment tested, NSOKE (10%) was found to be most effective in reducing the pest populations. The commercial formulation did not show any definite trend in reducing the sesamum pests. However, the most effective neem based commercial formulation in respect of Bihar hairy caterpillar, leaf webber and capsule borer and Sesamum Hawk moth was the nimbecidine but for sesamum gallfly, neemta.2100 was found to be most effective.

J. Taube, K. Vorkamp, M. Förster, R. Herrmann. (Department of Hydrology, University of Bayreuth, 95440, Bayreuth, Germany). Pesticide residues in biological waste. Chemosphere, 49(10) (2002), 1357-1365.

The aim of this study was to detect crop protection products (CPP) in single fractions of biological waste and to estimate pesticide concentrations in biological waste, depending on its composition. The composition of biological waste was determined from the literature, which provided information on its local and seasonal variability. The residue analysis included 17 pesticides for the analysis of tropical fruit peels and 48 pesticides for other fractions, e.g. vegetables, other fruits and ornamental flowers. The freeze-dried samples were extracted ultrasonically and purified by means of gel permeation chromatography. The analysis was carried out using gas chromatography with a mass selective detector in the selected ion monitoring mode. All tropical fruit peels contained CPP-residues. Thiabendazole was the most important contaminant with respect to the concentration and frequency of occurrence. The vegetables, other fruits and flowers also contained CPP-residues, such as the fungicides captan and dodemorph and the insecticide endosulfan. Based on import data and the composition of biological waste, the CPP-content of biological waste was estimated for rural and urban areas in summer and winter. Due to the higher percentage of tropical fruit peels, thiabendazole was the main contaminant in winter. The total concentration of CPP, summed up over all substances, ranged between 828 and 1829 $\mu\text{g (kg ww)}^{-1}$.

Kaushik N. Integrated pest management: status and concerns. In Innovative Pest and Disease Management in Horticultural and Plantation Crops, (2000), 129-130.

Crop species are infested by many insects and pathogens resulting in heavy yield losses amounting to Rs. 24,000 crores per annum. More than 100,000 microbes, 10,000 species of insects, 1000 nematode species and 30,000 weed species are found to cause damage to crop plants (1). The advent of synthetic insecticide specially DDT, Organophosphates, carbamates and herbicides like 2,4-D salt, etc during early-40s proved very effective in controlling the pest menace. This tempted the ignorant farmers to overuse and misuse the pesticides leading to unappeasable situation giving rise to problems in the form of insect resistance and resurgence, pesticide residues and ecological imbalances. The gravity of the problem can be judged by the fact that the DDT residues have entered in mother's milk through food chain (2) and pest-resurgence and resistance led to suicides by farmers in Andhra Pradesh, Maharashtra, Karnataka and Punjab. Realization of the ecological imbalances and long term impacts of the overuse of the pesticides favoured the emergence of Integrated Pest Management (IPM) during late sixties and early seventies. During the initial period, the concept of IPM focused on reducing the amount of pesticide by minimizing the number of sprays only. Later on integration of other methods of pest control became the major component of IPM while pesticide also remained a part of it.

Prem Kishore, A. K. Dikshit and Rajendra Kumar. (Division of Agricultural Chemicals, Indian Agricultural Research Institute, New Delhi-110012, India). Presence and bioefficacy of imidacloprid on sorghum and pearl millet. *Pestology*, 27(11) (2003), 6-9.

Sorghum and pearl millet seeds were treated with imidacloprid (Gaucho 70 WS, 7 and 14 g a.i. kg⁻¹ seed i.e. 10 and 20 g Gaucho 70 WS kg⁻¹ seed and Gaucho 600 FS, 7.2 and 14.4 g a.i. kg⁻¹ seed i.e. 12 and 24 g Gaucho 600 FS kg⁻¹ seed and sown in the field. The experiments were carried out in the field for three years to study the residues of imidacloprid (from both the formulations, 70 WS and 600 FS) in fodder and grains, stalks and soils at harvest from sorghum and pearl millet crops. Residues of imidacloprid were non-detectable in sorghum fodder from recommended dose (X) of both the formulations after 45 days of sowing, while in the case of pearl millet residues in fodder ranged ND to 0.12 and ND to 0.10 mg kg⁻¹ from 70 WS and 600 FS formulations, respectively. Residues were almost non-significant in both the fodders from double the recommended dose (2X). Matured grains of sorghum and pearl millet, their stalks and soil collected after harvest of the crops did not show the residues of imidacloprid from both the formulations. Both the formulations (70 WS and 600 FS) gave effective control of shoot fly in both the crops. The 4 X doses of imidacloprid i.e. 28 g a.i. kg⁻¹ seed (70 WS formulations) and 28.8 g a.i. kg⁻¹ seed (600 FS formulation) did not exhibit any noticeable phytotoxicity and adverse effect on the crops. Therefore, the X and 2X doses from both the formulations of imidacloprid involving seed treatments of sorghum and pearl millet were found safe and can find suitability in plant protection.

R. Nandakumar, S. Babu, R. Viswanathan, J. Sheela, T. Raguchander, R. Samiyappan. (Department of Plant Pathology, Tamil Nadu Agricultural University, Coimbatore 641003, India. Sugarcane Breeding Institute, Coimbatore 641007, India. Agricultural Research Station, Aliyar Nagar, India). A new bio-formulation containing plant growth promoting rhizobacterial mixture for the management of sheath blight and enhanced grain yield in rice. *BioControl*, 46(4) (2001), 493-510.

Three plant growth promoting rhizobacterial (PGPR) strains, PF1, FP7 and PB2, were tested alone and in combinations for suppression of rice sheath blight disease and promotion of plant growth under glasshouse and field conditions. The mixture of PGPR strains significantly reduced the sheath blight incidence when applied as either bacterial suspension through seed, root, foliar and soil application in glasshouse conditions, or as talc-based formulation under field conditions, compared to the respective individual strains. The

average mean of disease reduction was 29.2% for single strains and 45.1% for mixtures. In addition to disease suppression, treatment with mixture of PGPR strains promoted plant growth in terms of increased plant height and number of tillers, and ultimately grain yield. The average increases in yield for single strains were 17.7%, and 25.9% in case of mixture. Mixture of three PGPR strains reduced disease and promoted growth to a level equivalent to two strain mixtures. Though seed treatment of either single strain or strain mixtures alone could reduce the disease, subsequent application to root, leaves or soil further reduced the disease and enhanced the plant growth. The mixture consisting of PF1 plus FP7 was the most effective in reducing the disease and in promoting plant growth and grain yield.

S. Janarthanan, P. Suresh. Insecticidal potential of wild bean seed protein, arcelin. Natural product radiance, 2(5), 243-248.

During the co-evolutionary process, plants have yielded many organic compounds. These compounds have been found to overcome the predatory potential of herbivores. The use of such compounds in Integrated Pest Management (IPM) is on the rise. Wild pulses such as *Vigna* spp., *Cajanus* spp. and *Lablab* spp. found in Palney Hills, Western Ghats, Tamil Nadu were evaluated and reported to possess several such insect-antimetabolic compounds, of which arcelins play a major role in plant defense mechanism against their insect pests, mainly to bruchids (Osborn et al, 1988; Dillen et al, 1997; Goossens et al, 1999; Janarthanan et al, 2002). Arcelin has been identified and isolated for the first time from seeds of the wild specimens of *Lablab purpureus* (Linn.) Sweet in India and its antibiosis against the stored product insect pest, *Callosobruchus maculatus* (F.) is reported here.

S. K. Singh, A. K. Dikshit, K. D. Srivastava, R. S. Tanwar, P. Dureja. (Division of Plant Pathology, Division of Agricultural Chemicals, Indian Agricultural Research Institute, New Delhi-110012, India). Evaluation of Tebuconazole: Bioefficacy against spot blotch of wheat and residue studies. Pestology, 27(8) (2003), 13-17.

Efficacy of tebuconazole (Folicur 250 EW) against spot blotch (*Bipolaris sorokiniana*) of wheat was evaluated as spray treatment @ 0.05 and 0.1 percent of the formulate product, Folicur 250 EW (i.e. 0.0125 and 0.025% tebuconazole). Pre-inoculation treatment of the fungicide @ 0.1% reduced the disease severity from 38.6% to 5.1% (2000-01) and 36.3 to 6.3%(2001-02) under field conditions. The fungicide at this rate proved at par with propiconazole and better than triademefon and mancozeb. In addition to significant enhancement in yield, the residue analysis of the chemical did not reveal any residue in seed, soil and wheat straw at harvest.

Bioremediation

A. C. Silva, M. Dezotti and G. L. Sant'Anna, Jr. (Programa de Engenharia Quimica, COPPE/Universidade Federal do Rio de Janeiro, P.O. Box 68592, CEP 21945-970, Rio de Janeiro, Brazil). Treatment and detoxification of a sanitary landfill leachate. Chemosphere, 55(2) (2004), 207-214.

The leachate from an old sanitary landfill (Gramacho Metropolitan Landfill, Rio de Janeiro) was characterized and submitted to coagulation and flocculation treatment followed by ozonation and ammonia stripping. The performance of the treatment was assessed by monitoring the removal of organic matter (COD and TOC), ammonium nitrogen and metals. Detoxification was assessed by determining acute toxicity, using the following organisms: *Vibrio fisheri*, *Daphnia similis*, *Artemia salina* and *Brachydanio rerio*. Membrane fractionation was employed to infer the range of molecular masses of the pollutants found in the effluent, as well as the toxicity associated to these fractions. Of the techniques under investigation, coagulation and flocculation followed by ammonia stripping were the most effective for

toxicity and ammonium nitrogen removal. Membrane fractionation was effective for COD removal; however, acute toxicity was almost the same in all the fractionated samples. Ozonation was moderately effective for COD removal, but significant toxicity removal was only attained when high ozone doses were used.

A. D'Annibale, R. Casa, F. Pieruccetti, M. Ricci, R. Marabottini. (Dipartimento di Agrobiologia and Agrochimica, Università della Tuscia, Via San Camillo de Lellis snc, 01100, Viterbo, Italy. Dipartimento di Produzione Vegetale, Università degli Studi della Tuscia, Via San Camillo de Lellis snc, 01100, Viterbo, Italy). *Lentinula edodes* removes phenols from olive-mill wastewater: impact on durum wheat (*Triticum durum* Desf.) germinability. *Chemosphere*, 54(7) (2004), 887-894.

Olive-mill wastewater (OMW) exhibits highly phytotoxic properties, mainly due to phenols. A valuable option for OMW disposal is its agricultural use provided that phytotoxic effects are removed. The present investigation was aimed at evaluating the efficacy of the lignin-degrading fungus *Lentinula edodes* in achieving OMW detoxification. Germinability experiments on durum wheat showed that OMW phytotoxicity was significantly reduced by *L. edodes* cultures. Germinability on undiluted and twofold diluted OMW from fungal cultures was $34 \pm 5\%$ and $57 \pm 6\%$, respectively, while on related incubation controls it was almost completely suppressed. These results suggest that fungal cultures of *L. edodes* would decrease the phytotoxicity of this waste.

Araceli Linares, Juan Manuel Caba, Francisco Ligeró, Teresa de la Rubia, José artínez. (Departamento de Microbiología, Facultad de Farmacia, Universidad de Granada, Campus Cartuja, 18071, Granada, Spain. Departamento de Biología Vegetal, Facultad de Farmacia, Universidad de Granada, 18071, Granada, Spain). Detoxification of semisolid olive-mill wastes and pine-chip mixtures using *Phanerochaete flavido-alba*. *Chemosphere*, 51(9) (2003), 887-891.

Semisolid olive-mill residues, pine chips, and mixtures of both residues contain phytotoxic components capable of inhibiting germination and vegetative growth in plants. Solid-state cultures of *Phanerochaete flavido-alba* on pine chips or mixtures of both residues reduce these phytotoxic effects in fermented substrates. The phenol and lipid contents in cultures detoxified by this fungus also decreases.

C. Quintelas, T. Tavares. (Centro de Engenharia Biológica – IBQF, Universidade do Minho, 4710-057 Braga, Portugal). Removal of chromium(VI) and cadmium(II) from aqueous solution by a bacterial biofilm supported on granular activated carbon. *Biotechnology Letters*, 23(16) (2001), 1349-1353.

A biofilm of *Arthrobacter viscosus*, supported on granular activated carbon, removed between 100% and 50% of Cr(VI) and between 100% and 20% of Cd(II) from solutions with initial concentrations between 4–11 mgmetal l⁻¹ and a flow residence time of 1.2 min. For experiments of lower initial concentrations, a steady-state removal of 50% was reached after 71 bed volumes of Cr solution passed through the biosorbent bed and a steady-state removal of 30% was reached after 47 bed volumes of Cd solution passed through a similar bed. Final uptakes of 8.5 mgCr gcarbon⁻¹ and 4.2 mgcd gcarbon⁻¹ were determined for initial concentrations of 10 mgCr l⁻¹ and 11 mgCd l⁻¹, respectively. The influence on the overall process of two different surface treatments of the support was evaluated and compared with the behavior of a support not treated.

Carlo Viti, Alessandra Pace, Luciana Giovannetti. (DiBA, sez. Microbiologia, Università degli Studi di Firenze, P.le delle Cascine 24, I 50144 Firenze, Italy). Characterization of Cr(VI)-Resistant Bacteria Isolated from Chromium-Contaminated Soil by Tannery Activity. *Current Microbiology*, 46(1) (2003), 1 – 5.

Bacterial strains, previously isolated from a chromium-polluted soil, were identified on the basis of Gram reaction and biochemical characteristics (Biolog system). Moreover, chromate MICs, chromate reduction capability, multiple heavy metal tolerance, and antibiotic susceptibility were tested for each isolate. All strains but one were Gram-positive and resistant to high concentrations of chromate. The most Cr(VI)-resistant isolate (22mM) was identified as *Corynebacterium hoagii*. All Cr(VI)-resistant strains except the isolate ChrC20 were capable of catalyzing the reduction of Cr(VI) to Cr(III), a less toxic and less water-soluble form of chromium. The only isolate Cr(VI)-sensitive, attributed to the *Pseudomonas* genus, also exhibited Cr(VI)-reduction. Isolates were also screened for the presence of plasmid DNA. The strains ChrC20 and ChrB20 harbored one and two plasmids of high molecular mass, respectively. This approach permitted selection of some bacterial strains, which could be used for bioremediation of Cr(VI)-polluted environments.

Dong W. Kim, Daniel K. Cha, J. Wang, C. P. Huang. (Department of Civil & Environmental Engineering, University of Delaware, Newark, DE 19711, USA). Heavy metal removal by activated sludge: influence of *Nocardia amarae*. *Chemosphere*, 46(1) (2002), 137-142.

The goal of this research was to examine the metal binding capacity of *Nocardia amarae* cells and to assess the influence of *Nocardia* cells on the overall metal binding capacity of activated sludge. Metal sorption capacities of the pure *Nocardia* cells and activated sludge biomass containing various levels of added *Nocardia* pure cultures were determined by a series of batch experiments. Batch sorption isotherms for nickel (Ni), copper (Cu), and cadmium (Cd) showed that the pure culture of *N. amarae* exhibited significantly higher metal sorption capacity than the activated sludge biomass obtained from Wilmington Wastewater Treatment Plant (Wilmington, DE). Surface area of biomass estimated by a dye technique showed that pure *N. amarae* cells growing at stationary phase have substantially more specific surface area than that of activated sludge from Wilmington Treatment Plant. A two-fold difference in specific surface area indicated that the higher metal sorption capacity of *Nocardia* cells may be due to the higher specific surface area. The metal sorption capacity of activated sludge increased proportionally with the amount of *Nocardia* cells present in the mixed liquor. This increase was attributed to the greater specific surface area of the mixed liquor samples containing greater amounts of *Nocardia* cells.

Geremias R., Pedrosa R.C., Benassi J.C., Fávere V.T., Stolberg J., Menezes C.T.B., Laranjeira M.C.M. Remediation of Coal Mining Wastewaters using Chitosan Microspheres. *Environmental Technology*, 24(12) (2003), 1509-1515.

This study aimed to evaluate the potential use of chitosan and chitosan/poly(vinylalcohol) microspheres incorporating with tetrasulphonated copper (II) phthalocyanine (CTS/PVA/TCP) in the remediation of coal mining wastewaters. The process was monitored by toxicity tests both before and after adsorption treatments with chitosan and microspheres. Physico-chemical parameters, including pH and trace-metal concentration, as well as bioindicators of water pollution were used to that end. Wastewater samples collected from drainage of underground coal mines, decantation pools, and contaminated rivers were scrutinized. Acute toxicity tests were performed using the Brine Shrimp Test (BST) in order to evaluate the remediation efficiency of different treatments. The results showed that the pH of treated wastewater samples were improved to values close to neutrality. Chitosan treatments were also effective in removing trace-metals. Pre-treatment with chitosan followed by microsphere treatment (CTS/PVA/TCP) was more effective in decreasing toxicity than the treatment using only chitosan. This was probably due to the elimination of pollutants other than trace-metals. Thus, the use of chitosan and microspheres is an adequate alternative towards remediation of water pollution from coal mining.

Hilloi Guha, Krishnaswamy Jayachandran, Florentin Maurrasse. (Miami-Dade County, Department of Environmental Resources Management, 33 SW 2nd Avenue, PH-2, Miami, FL 33130, USA. Environmental Studies Department and Southeast Environmental Research Center, Florida International University, Miami, FL 33199, USA. Department of Earth Science, Florida International University, Miami, FL 33199, USA). Microbiological reduction of chromium(VI) in presence of pyrolusite-coated sand by *Shewanella alga* Simidu ATCC 55627 in laboratory column experiments. *Chemosphere*, 52(1) (2003), 175-183.

Hexavalent chromium (Cr(VI)) was reduced to non-toxic trivalent chromium (Cr(III)) by a dissimilatory metal reducing bacteria, *Shewanella alga* Simidu (BrY-MT) ATCC 55627. A series of dynamic column experiments were conducted to provide an understanding of Cr(VI) reduction by the facultative anaerobe BrY-MT in the presence of pyrolusite (β -MnO₂) coated sand and uncoated-quartz sand. All dynamic column experiments were conducted under growth conditions using Cr(VI) as the terminal electron acceptor and lactate as the electron donor and energy source. Reduction of Cr(VI) was rapid (within 8 h) in columns packed with uncoated quartz sand and BrY-MT, whereas Cr(VI) reduction by BrY-MT was delayed (57 h) in the presence of β -MnO₂-coated sand. The role of β -MnO₂ in this study was to provide oxidation of trivalent chromium (Cr(III)). BrY-MT attachment was higher on β -MnO₂-coated sand than on uncoated quartz sand at 10, 60, and 85.5 h. Results have shown that this particular strain of *Shewanella* did not appreciably reduce Mn(IV) to Mn(II) species nor biosorbed Cr and Mn during its metabolic activities.

K Murugesan. Bioremediation of paper and pulp mill effluents. *Indian Journal of Experimental Biology*, 41 (2003), 1239-1248.

Pulp and paper mill effluents pollute water, air and soil, causing a major threat to the environment. Several methods have been attempted by various researchers throughout the world for the removal of colour from pulp and paper mill effluents. The biological colour removal process uses several classes of microorganisms – bacteria, algae and fungi – to degrade the polymeric lignin derived chromophoric material. White rot fungi such as *Phanerochaete chrysosporium*, *Coriaria versicolor*, *Trametes versicolor* etc., are efficient in decolourizing paper and pulp mill effluents. *Gliocladium virens*, a saprophytic soil fungus decolourised paper and pulp mill effluents by 42% due to the production of hemicellulase, lignin peroxidase, manganese peroxidase and laccase.

Kelly P. Nevin, Kevin T. Finneran, Derek R. Lovley. (Department of Microbiology, University of Massachusetts, Amherst, Massachusetts 01003). Microorganisms Associated with Uranium Bioremediation in a High-Salinity Subsurface Sediment. *Applied and Environmental Microbiology*, 69(6) (2003), 3672-3675.

Although stimulation of dissimilatory metal reduction to promote the reductive precipitation of uranium has been shown to successfully remove uranium from some aquifer sediments, the organisms in the family Geobacteraceae that have been found to be associated with metal reduction in previous studies are not known to grow at the high salinities found in some uranium-contaminated groundwaters. Studies with a highly saline uranium-contaminated aquifer sediment demonstrated that the addition of acetate could stimulate the removal of U(VI) from the groundwater. This removal was associated with an enrichment in microorganisms most closely related to *Pseudomonas* and *Desulfosporosinus* species.

Lal B, Mishra S, Ramesh KC Jyot J. Oilzapper: an effective bioremediation tool. *Proceedings of the international Seminar on Oily Sludge Management*, (2000), 1-2.

Oil refineries generate large quantities of oily sludge, the safe disposal of which is major problem. The sludge is generated during cleaning of storage tanks, cleaning and desilting of oil separator basins, distillation column residues, exchanger tube bundle sludge and sludge generated from effluent treatment plant (ETP). Conventional disposal method involves storing in sludge pits, which are expensive to construct and add to the already limited land resource at the refinery. Furthermore possible seepage of the constituents into groundwater can't be ruled out. An alternative method of oily sludge management involves use of microorganisms for in situ degradation in the soil. The process called Bioremediation is eco-friendly and more cost effective compared to conventional methods. Oilzapper, a consortium of oily sludge degrading bacteria, was developed in our laboratory to expedite the rate of bioremediation in contaminated soil. Extensive studies under laboratory conditions had established the potential of oilzapper to degrade oily sludge and crude oil. Field study was carried out in a refinery to evaluate the credibility of using Oilzapper to reclaim oily sludge contaminated land. A 4000 m² of land (site A) at Barauni refinery was taken up for the present study. The study was carried out for a period of one year from May 1998 to May 1999. A total of 900 tonnes of different types of oily sludge was loaded onto the site. Oilzapper at the rate of 1 kg/10 m² of contaminated land was applied. Out of a total of 300 tonnes of ETP sludge added from May 98-September 98 around 90.2-96% biodegradation was noted (adjacent figure-shows the representative data). The figure clearly suggests the effectiveness of oilzapper for reclamation of the contaminated land. In September 98, 200 tonnes of tank bottom sludge was loaded. Total biodegradation between 37.7- 66.4% was noted during the next 60 days. Residual oily sludge was added for the third (100 tonnes) and fourth (300 tonnes) sludge loading was added onto the site. Rates of biodegradation varied from 53.7-85.4% during the next 120 days till the end of the study. Initial results (May 98-December 98) encouraged us to take up another 2000m² of land (site B). The study was carried out for 150 days till May 99. Around 1500 tonnes of tank bottom sludge was loaded onto this field and treated with oilzapper. An initial load of 24.9% sludge was calculated at the beginning of the study. At the end of the study the contamination load was 3.63% which accounted for a reduction by 85.4%. In both the sites a small area was demarcated as control where oilzapper was not applied. The reduction of oily sludge in the untreated plots was found to be very less in comparison to the treated plots. The study established oilzapper as a potential bioremediation tool, which accounted for effective reduction of 2400 tonnes of oily sludge loaded onto both the sites.

Lance D. Hansen, Cathy Nestler, Dave Ringelberg, Rakesh Bajpai. (USACE Engineer Research and Development Center, 3909 Halls Ferry Road, Vicksburg, MS, USA. Applied Research Associates, Inc., 119 Monument Place, Vicksburg, MS, USA. USACE Cold Regions Research Engineer Laboratory, Hanover, NH, USA. Department of Chemical Engineering, University of Missouri, Engineering Building East, W2016, Columbia, MO 65211, USA). Extended bioremediation of PAH/PCP contaminated soils from the POPILE wood treatment facility. Chemosphere, 54(10) (2004), 1481-1493.

A study was conducted using two pilot-scale land-treatment units (LTUs) to evaluate the efficacy of different cultivation and maintenance schedules during bioremediation of contaminated soil from a wood treatment facility using landfarming technology. The soil contained high concentrations of polycyclic aromatic hydrocarbons (PAHs, ~13 000 ppm) as well as of pentachlorophenol (PCP, ~1500 ppm). An initial 6-month intensive-treatment phase was followed by 24 months of less-intensive treatment. During the first phase, traditional landfarming practice of regular cultivation was compared with a gas-phase composition based cultivation strategy, and both the landfarming units were intensively monitored and maintained with respect to moisture control and delivery of nutrients. The two strategies resulted in similar contaminant concentration profiles with time during this

phase, although different microbial populations developed in the two-landfarming units. The second (less-intensive) treatment phase involved no moisture control and nutrient delivery beyond the initial adjustments, and compared natural attenuation (no cultivation) with quarterly cultivation of soil. Both the strategies showed similar behavior again. GC/MS analysis of the soil samples showed PAH removal including four-ring homologues. Leachability tests at zero time and after 6 and 22 months of operation showed significant reductions in leaching of PCP and low molecular weight PAHs. Extended treatment resulted in some leaching of high molecular weight PAHs. Significant biological activity was demonstrated, even at the high contaminant concentrations. Phospholipid ester-linked fatty acid (PLFA) analysis showed an increase in biomass and a divergence in community composition in soils depending on the treatment conducted.

Le Floch S., Guyomarch J., Merlin F., Børseth J.F., Le Corre P., Lee K. Effects of Oil and Bioremediation on Mussel (*Mytilus edulis* L.) Growth in Mudflats. *Environmental Technology*, 24(10) (2003), 1211-1219.

Mussels (*Mytilus edulis* L.) were exposed to crude oil during a field experiment to evaluate two bioremediation strategies (nutrient addition and nutrient addition with tilling). The mussels were placed in 4 mesocosms: Control, Oil, Oil + Nutrients, and Oil + Nutrients + Tilled. Tilling appeared to be clearly detrimental to mussel growth. Additionally, this field experiment demonstrated that at temperatures below 5°C, growth was reduced to rates undetectable by the laser diffraction method. The data on mussel shell length show that this technique does offer very sensitive and useful comparative measurements of physiological function. Measurement of shell growth has the advantage over other techniques in that it is non-invasive and non-destructive and thus may be used continuously without disturbing critical physiological and biochemical functions; however, bivalve physiology is strongly linked to environmental conditions, so it is important to include such measures (i.e. seawater temperature and turbidity) in the design of the biomonitoring program. Elevated polycyclic aromatic hydrocarbon (PAH) levels reflected bioaccumulation in mussels from all the oiled mesocosms. This correlated with reduction in growth rate. Maximum reduction in growth was observed in mussels from the tilled mesocosm which contained the lowest phenanthrene and dibenzothiophene concentrations. The tilling caused an increase in suspended solids which inhibited filter feeding activity, and resulted in suppressed growth and slower intake of PAH-laden sediment.

M. Humar, M. Bokan, S. A. Amartey, M. entjurc, P. Kalan, F. Pohleven. (Department of Wood Science and Technology, Biotechnical Faculty, University of Ljubljana, Ljubljana, Slovenia. Forest Products Research Center, Buckinghamshire Chilterns, University College, High Wycombe, UK. Institute Jozef Stefan, Ljubljana, Slovenia. Slovenian Forestry Institute, Ljubljana, Slovenia). Fungal bioremediation of copper, chromium and boron treated wood as studied by electron paramagnetic resonance. *International Biodeterioration & Biodegradation*, 53(1) (2004), 25-32.

In future years, problems concerning the disposal of waste copper/chromium-treated wood will increase significantly. One of the environmentally friendly options of dealing with such treated wood is through bioremediation with copper-tolerant wood decay fungi in order to recycle both the wood fibers and the heavy metals. To study changes during the bioremediation process, Norway spruce (*Picea abies*) samples were vacuum impregnated with 5% CCB solution. Some samples were also impregnated with copper or chromium solution of the same concentration as in the CCB preservative. Following conditioning of the samples, they were then exposed to two copper-tolerant brown rot fungi, (*Antrodia vaillantii*, *Leucogyrophana pinastri*) and two copper-sensitive brown rot fungi, (*Gloeophyllum trabeum*, *Poria monticola*) for a period of 4–8 weeks. After exposure, the

samples were cleaned of the mycelia and leached with water or 1.25% ammonia solution for 4 days. The concentrations of Cr and Cu in the leachates were determined. After the leaching process, the samples were studied using electron paramagnetic resonance (EPR). The results obtained showed the important role oxalic acid produced by the decay fungi plays during leaching of the metals from the treated wood. Furthermore, it was also found that though excretion of oxalic acid is necessary for the leaching of metals, it does not fully explain fungal ability to decay copper preserved wood.

M.R. Kosseva, C.A. Kent, D.R. Lloyd. (Centre for Biochemical Engineering, School of Chemical Engineering, The University of Birmingham, Edgbaston B15 2TT Birmingham, UK). Thermophilic bioremediation of whey: effect of physico-chemical parameters on the efficiency of the process. Biotechnology Letters, 23(20) (2001), 1675-1679.

A two-stage process for the bioremediation of blue Stilton whey has been developed. It employs both naturally occurring thermotolerant organisms found in whey (lactic acid bacteria and yeast) and a thermophilic isolate (*Bacillus* sp.). Thermophilic digestion occurred only at neutral pH. Multiple substrates were consumed simultaneously under mesophilic but not thermophilic conditions.

Mishra S, Jyot J, Kuhad R C, Lal B. Evaluation of inoculum addition to stimulate in situ bioremediation of oily sludge Contaminated soil. Applied and Environmental Microbiology, (2001), 1675-1781.

Oily sludge/crude oil contamination is a major environmental concern since of the constituent hydrocarbons are toxic. Besides this incidence of crude oil spill is increasing despite the best effort of petro-chemical industry. Natural diversity of soil microorganisms is a useful source for detoxification of hazardous waste contamination sites. Understanding of the microbial diversity in such an ecosystem will lead to formulation of better strategies of bioremediation. The study was taken up to estimate the diversity of hydrocarbon degrading organisms. Molecular typing of hydrocarbon degrading bacterial strains was performed to study the genetic diversity among the isolated bacterial strains. In a full-scale bioremediation study plot A and plot B were treated with a bacterial consortium and nutrients, which resulted in 92.0% and 89.7% removal of TPH respectively in one year as compared to 14.0% removal of TPH in the control plot C. In plot A the alkane fraction of TPH was reduced by 94.2%, the aromatic fraction of TPH for 9.1.9% and NSO (nitrogen, sulfur, oxygen containing compound) and asphaltene fractions of TPH were removed 85.2% in one year. Similarly in plot B degradation of alkane, aromatic and asphaltene, NSO fractions of TPH were 95.1% 94.8% and 63.5% respectively in 345 days. However in control plot C removal of alkane (17.3%), aromatic (12.9%), NSO plus asphaltene fractions (5.8%) was much less. The population of introduced *Acinetobacter baumannii* strains in plots A and B was stable even after one year of its application. Physical and chemical properties of the soil at the bioremediation site improved significantly in one year.

Nirmala Agrawal, R.S. Sachan. (Department of Soil Science, College of Agriculture, G.B. Pant University of Agriculture and Technology, Pantnagar-263145, Uttaranchal, India). Physicochemical characterization of the wastes of sugar and yeast industries. Poll Res. 22(4) (2003), 585-589.

The disposal of industrial effluents and solid wastes is a problem of increasing importance throughout the world. A huge amount of solid waste and waste waters are generated from sugar factories and molasses based yeast-producing factories. The sugar industry effluent was characterized by pH (5.64), EC ($.794 \text{ dSm}^{-1}$) and high COD (1400 mg/L). This waste water contained a considerable amount of plant nutrients viz. N, P, K, Ca, Mg, S and micronutrients. The yeast industry effluent was characterized by high pH (7.09), EC (6.60),

BOD (869.5 mg/L) and COD (15000 mg/L). This wastewater also had considerable amounts of plant nutrients and their contents were higher as compared to sugar industry effluent. Likewise the solid wastes of sugar and yeast industries also tested considerable amounts of recyclable macro and micronutrients though contained high amount of organic carbon.

Nirupama Mallick. (Agricultural and Food Engineering Department, Indian Institute of Technology, Kharagpur-721 302, India). Biotechnological potential of immobilized algae for wastewater N, P and metal removal: A review. *BioMetals*, 15(4) (2002), 377-390.

This presentation comprises a review on the use of immobilized algae for wastewater nitrogen, phosphorus and metal removal purposes. Details of the use of immobilized algae, the techniques of immobilization and the effects of immobilization on cell function are included. Particularly relevant in their use for heavy metal removal from wastewaters; upon enriching the biomass in metal, can be recovered, thereby providing economic advantages. The use of immobilized microalgae in these processes is very adequate and offers significant advantages in bioreactors. The future of this area of algal cell biotechnology is considered.

Olivier Potin , Catherine Rafin, Etienne Veignie. (Université du Littoral-Côte d'Opale, 50 rue Ferdinand Buisson, BP 699, 62228, Calais Cédex, France). Bioremediation of an aged polycyclic aromatic hydrocarbons (PAHs)-contaminated soil by filamentous fungi isolated from the soil. *International Biodeterioration & Biodegradation*, 54(1) (2004), 45-52.

Twenty-one filamentous fungi were isolated from the soil of an old polycyclic aromatic hydrocarbon (PAH)-contaminated gaswork site and tested in their native soil for PAH degradation. This degradation study was performed for each isolate with two inoculation treatments, by spore or mycelial inoculum. An improvement in the extent of total PAH degradation occurred with mycelial inoculum. The greatest degradation was obtained with *Coniothyrium* sp. (26.5%) and *Fusarium* sp. (27.5%) inoculum, especially for PAHs of high-molecular-weight that contained more than three fused aromatic rings. Correlations between mycobiota capacity (in treatments inoculated with mycelium) and PAH characteristics (structure, water solubility, bioavailability) were obtained from correspondence factorial analysis. This study suggests that such filamentous fungi could be used in clean-up of long-term contamination of soils by PAHs.

Omry Koren, Vishnia Knezevic, Eliora Z. Ron, Eugene Rosenberg. (Department of Molecular Microbiology and Biotechnology, Tel Aviv University, Ramat Aviv, Israel 69978). Petroleum Pollution Bioremediation Using Water-Insoluble Uric Acid as the Nitrogen Source. *Applied and Environmental Microbiology*, 69(10) (2003), 6337-6339.

The biodegradation of hydrocarbon pollutants in open systems is limited by the availability of a utilizable nitrogen source. This limitation can be overcome by using uric acid. Enrichment cultures grown on crude oil-uric acid media yielded mixed and pure cultures that degraded petroleum. In a simulated open system, uric acid bound to crude oil and was available for bacterial growth and petroleum biodegradation.

R. Bennicelli, Z. Stępniewska, A. Banach, K. Szajnocha, J. Ostrowski. (Department of Biochemistry and Environmental Chemistry, Catholic University of Lublin, Al. Kraśnicka 102, 20-718, Lublin, Poland. Institute of Agrophysics PAS, Doświadczalna 4, 20-290, Lublin 27, Poland. WIOŚ, Obywatelska 13, 20-092, Lublin, Poland). The ability of *Azolla caroliniana* to remove heavy metals (Hg(II), Cr(III), Cr(VI)) from municipal waste water. *Chemosphere*, 55(1) (2004), 141-146.

The aim of this paper was to investigate the capacity of a small water fern, *Azolla caroliniana* Willd. (Azollaceae), to purify waters polluted by Hg and Cr. Many plants are capable of accumulating heavy metals (called hyperaccumulators) and one of them is the water fern *A. caroliniana*. During 12 days of the experiment the fern was grown on the nutrient solution containing Hg^{2+} , Cr^{3+} and CrO_4^{2-} ions, each in a concentration 0.1, 0.5 and 1.0 mg dm^{-3} . The presence of these ions caused a 20–31% inhibition of *A. caroliniana* growth, the highest in the presence of Hg(II) ions, in comparison to the control. After day 12 of the experiment, metal contents the solution decreased to 0–0.25 mg dm^{-3} , and this decrease comprised between 74 (Cr^{3+} 1.0 mg dm^{-3} treatment) and 100% (CrO_4^{2-} 0.1 mg dm^{-3} treatment). The fern took a lesser quantity of the metals from 0.1 mg dm^{-3} treatments compared to 0.5 and 1.0 mg dm^{-3} treatments. In the *A. caroliniana* tissues the concentration of heavy metals under investigation ranged from 71 to $964 \text{ mg kg}^{-1} \text{ dm}$; the highest level being found for Cr(III) containing nutrient solution.

Riina Turpeinen, Timo Kairesalo, Max M. Häggblom. (Department of Ecological and Environmental Sciences, University of Helsinki, Lahti, Finland. Department of Biochemistry and Microbiology, Rutgers, The State University of New Jersey, New Brunswick, NJ 08901-8525, USA). Microbial community structure and activity in arsenic-, chromium- and copper-contaminated soils. FEMS Microbiology Ecology, 47(1) (2004), 39-50.

Microbial community structure, potential microbial activity and As resistance were affected by arsenic (As), chromium (Cr) and copper (Cu) contamination in soils of abandoned wood impregnating plants. Contaminated soils differed in the concentrations of soil acid-soluble and total water-soluble As, Cr and Cu, and in the concentration of bioavailable As analyzed with a bacterial sensor. Phospholipid fatty acid (PLFA) and 16S rRNA gene terminal restriction fragment length polymorphism (t-RFLP) profiles indicated that exposure to high metal contamination or subsequent effects of this exposure permanently changed microbial community structure. The total number of colony forming units (CFU) was not affected by metal contamination and the As(V)-resistant bacterial ratio to total heterotrophic plate counts was high (0.5–1.1) and relatively independent of the concentration of As. In contrast, the proportion of As(III)-resistant bacteria was dependent on the concentration of As in the soils and a significant positive relationship was found between the bioavailability of As and the proportion of As(III)-resistant bacteria. Dominant As-resistant isolates from contaminated soils were identified by their fatty acid methyl ester (FAME) profiles as *Acinetobacter*, *Edwardsiella*, *Enterobacter*, *Pseudomonas*, *Salmonella* and *Serratia* species. No differences were noted in glucose mineralization among contaminated and control soil samples within sites. Based on [^{14}C]glucose mineralization the community was able to compensate for the reduced diversity. According to t-RFLP results, this was not due to a reversion towards the unexposed community, but mainly due to the appearance of new dominating species. This study, combining complementary culture-dependent and -independent methods, suggests that microbes are able to respond to soil metal contamination and maintain metabolic activity apparently through changes in microbial community structure and selection for resistance.

Shardendu, N. Salhani, S. F. Boulyga, E. Stengel. (Department of Botany and Vocational Centre of Biotechnology, Bihar National College, Patna University, Pin-800004, India. Institute of Chemistry and Dynamics of Geosphere, Institute for Phytosphere (ICG-III), Research Centre Juelich, D-52425, Juelich, Germany. Radiation Physics and Chemistry Problems Institute, Sosny, Minsk, Belarus). Phytoremediation of selenium by two helophyte species in subsurface flow constructed wetland. Chemosphere, 50(8) (2003), 967-973.

The phytoremediation of selenium by two different wetland species was investigated. Selenium (20.4 µg/l) was supplied continuously to subsurface flow constructed wetlands, one vegetated with *Typha latifolia* L. and the other with *Phragmites australis* (Cav.) Trin. ex Steud. The beds of both species had same hydraulic loading rate (0.079 m³/m²/d) and water retention time (24 h). However, the mass loading rate was 1.27 mg Se/m²/d for *Phragmites* and 1.35 mg Se/m²/d for *Typha*. In the *Typha* bed Se migrated faster than in the *Phragmites* bed. After 25 d of Se supplementation in the *Typha* bed about 54% of the Se inlet concentration remained in the outlet water. In the *Phragmites* bed Se was removed completely from the water after passing through 3/4 of the bed length. After 65 d of Se supplementation the highest amount of Se (2.8 µg/g dry matter) was determined in the organic material of the *Typha* bed. Roots and rhizomes accumulated 2.2 and 1.8 µg/g dry matter respectively. *Phragmites* accumulated Se in the leaves and stems, but not in the rhizomes. The accumulation in the leaves (1.8 µg Se/g dry matter) was three times higher than in the stems (0.6 µg Se/g dry matter).

Shen-Yi Chen, Jih-Gaw Lin. (Institute of Environmental Engineering, National Chiao Tung University, 75 Po-Ai Street, Hsinchu 300, Taiwan). Bioleaching of heavy metals from livestock sludge by indigenous sulfur-oxidizing bacteria: effects of sludge solids concentration. *Chemosphere*, 54(3) (2004), 283-289.

A technologically and economically feasible process called bioleaching was used for the removal of heavy metals from livestock sludge with indigenous sulfur-oxidizing bacteria in this study. The effects of sludge solids concentration on the bioleaching process were examined in a batch bioreactor. Due to the buffering capacity of sludge solids, the rates of pH reduction, ORP rise and metal solubilization were reduced with the increase of the solids concentration. No apparent influence of solids concentration on sulfate produced by sulfur-oxidizing bacteria was observed when the solids concentration was less than 4% (w/v). A Michaelis–Menten type of equation was able to well describe the relationship between solids concentration and rate of metal solubilization. Besides, high efficiencies of metal solubilization were achieved after 16 d of bioleaching. Therefore, the bioleaching process used in this study could be applied to remove heavy metals effectively from the livestock sludge.

Solange In[^]es Mussatto, In[^]es Conceição Roberto. (Department of Biotechnology, Faculty of Chemical Engineering of Lorena, Lorena-SP 12600-970, Brazil). Hydrolysate detoxification with activated charcoal for xylitol production by *Candida guilliermondii*. *Biotechnology Letters*, 23(20) (2001), 1681-1684.

A detoxification method using activated charcoal with concentrated rice straw hemicellulosic hydrolysate improved the conversion of xylose to xylitol by the yeast *Candida guilliermondii* by 22%. This was achieved when the hydrolysate:charcoal ratio was 40 g g⁻¹, resulting in removal of 27% of phenolic compounds. Under this condition, the xylitol yield factor (0.72 g g⁻¹) and volumetric productivity (0.61 g l⁻¹ h⁻¹) were close to those attained in a semi-defined medium simulating hydrolysate sugars.

Sugijanto Kartosentono, Ana Nuraida, Gunawan Indrayanto, Noor Cholies Zaini. (Laboratories of Pharmaceutical Biotechnology. Phytochemistry, Faculty of Pharmacy, Airlangga University, Jl. Dharmawangsa Dalam, Surabaya 60286, Indonesia). Phytoremediation of Sr²⁺ and its influence on the growth, Ca²⁺ and solasodine content of shoot cultures of *Solanum laciniatum*. *Biotechnology Letters*, 23(2) (2001), 153-155.

Shoot cultures of *Solanum laciniatum* were able to remediate Sr²⁺ contained in the media (25–200 mg l⁻¹) and to accumulate Sr²⁺ up to 0.13% in the biomass. The growth of these shoot cultures remained good. Sr²⁺ at 50 and 100 mg l⁻¹ caused the vascular system of

the stem to develop outwards with an increase of solasodine content 1.6 and 1.4 fold, respectively. A decrease of Ca²⁺ content of 16% was observed at Sr²⁺+100 and 200 mg l⁻¹.

Thomas Pümpel, Lynne E. Macaskie, John A. Finlay, Ludo Diels, Marios Tsezos. (Institut für Mikrobiologie, Universität Innsbruck, Technikerstrasse 25, A-6020 Innsbruck, Austria. School of Biosciences, University of Birmingham, Birmingham B15 2TT, UK. Vlaamse Instelling voor Technologisch Onderzoek (VITO), Boeretang 200, B-2400 Mol, Belgium. National Technical University of Athens, Heroon Polytechniou 9, GR-15780 Zografou, Greece). Nickel removal from nickel plating waste water using a biologically active moving-bed sand filter. *BioMetals*, 16(4) (2003), 567-581.

Efficient removal of dissolved nickel was observed in a biologically active moving-bed 'MERESAFIN' sand filter treating rinsing water from an electroless nickel plating plant. Although nickel is fully soluble in this waste water, its passage through the sand filter promoted rapid removal of approximately 1 mg Ni/l. The speciation of Ni in the waste water was modelled; the most probable precipitates forming under the conditions in the filter were predicted using PHREEQC. Analyses of the Ni-containing biosludge using chemical, electron microscopical and X-ray spectroscopic techniques confirmed crystallisation of nickel phosphate as arupite (Ni₃(PO₄)₂·8H₂O), together with hydroxyapatite within the bacterial biofilm on the filter sand grains. Biosorption contributed less than 1% of the overall sequestered nickel. Metabolising bacteria are essential for the process; the definitive role of specific components of the mixed population is undefined but the increase in pH promoted by metabolic activity of some microbial components is likely to promote nickel desolubilisation by others.

W. M. Law, W. N. Lau, K. L. Lo, L. M. Wai, S. W. Chiu. (Environmental Science Programme, The Chinese University of Hong Kong, Shatin, N.T., Hong Kong SAR, China. Department of Biology, The Chinese University of Hong Kong, Shatin, N.T., Hong Kong SAR, China). Removal of biocide pentachlorophenol in water system by the spent mushroom compost of *Pleurotus pulmonarius*. *Chemosphere*, 52(9) (2003), 1531-1537.

Pentachlorophenol (PCP) has been widely used as a wood preservative since 1980s. Although it has been banned worldwide, residues of PCP are still commonly found. The spent compost of oyster mushroom *Pleurotus pulmonarius* (SMC) which was a degraded paddy straw-based substrate, contained 25% chitin. Five percentage of the SMC could remove 89.0 ± 0.4% of 100 mg PCP l⁻¹ within 2 days at room temperature predominantly by biodegradation. The maximum removal capacity was 15.5 ± 1.0 mg g⁻¹ SMC. The sorption kinetics of PCP by SMC can be described by the Freundlich monolayer model with a theoretical sorption capacity similar to that found for chitin. A PCP-degradative bacterium was isolated from the SMC. Yet, biodegradation was predominantly contributed by the immobilized ligninolytic enzymes secreted by the mushroom to the SMC. Degradation of PCP involves dechlorination, methylation, carboxylation and ring cleavage as verified by GC-MSD and ion chromatography. Thus, the SMC has a potential for treating PCP-contaminated water.

Y. X. Chen, Q. Lin, Y. M. Luo, Y. F. He, S. J. Zhen, Y. L. Yu, G. M. Tian, M. H. Wong. (Department of Environmental Science, Zhejiang University, Huajiachi, Hangzhou 310029, China. Institute of Soil Science, Chinese Academy of Sciences, P.O. Box 821, Nanjing 210008, China. Institute for Natural Resources and Waste Management, Department of Biology, Hong Kong Baptist University, Kowloon, PR China). The role of citric acid on the phytoremediation of heavy metal contaminated soil. *Chemosphere*, 50(6) (2003), 807-811.

Adsorption and hydroponics experiments were conducted to study the role of citric acid on the phytoremediation of heavy metal contaminated soil. The results show that addition of citric acid decreased the adsorption of both lead and cadmium, such an effect was bigger for cadmium than for lead. The decrease in the adsorption of Pb and Cd was mainly due to a decrease of pH in the presence of citric acid. The presence of citric acid could alleviate the toxicity of Pb and Cd to radish, and stimulate their transportation from root to shoot. The studies of heavy metal forms using sequential extraction demonstrated that lead was mainly existed as F_{HAC} (a lower bioavailable form) in the root, while F_{HCl} was the dominant form in the leaf. The addition of citric acid to the soil changed the concentration and relative abundance of all the forms. The detoxifying effect of citric acid to Pb in shoots might result from the transformation of higher toxic forms into lower toxic forms. Cadmium was mainly present as F_{NaCl} , therefore, it had higher toxicity than lead. The addition of citric acid increased the abundance of $F_{H_2O}+F_{NaCl}$, indicating that citric acid treatment could transform cadmium into more transportable forms.

Biosensor

A. P. Soldatkin, J. Montoriol, W. Santc, C. Martelet, N. Jaffrezic-Renaulta. (Ecole Centrale de Lyon, UMR/CNRS 5621, 36 Av. Guy de Collongue, F69134, Ecully Cedex, France. Institute of Molecular Biology and Genetics, National Academy of Sciences of Ukraine, 150 Zabolotnogo Street, Kiev 03143, Ukraine. HEMODIA S.A., Rue du Chêne Vert, BP 148, 31676, Labège Cedex, France). Novel urea sensitive biosensor with extended dynamic range based on recombinant urease and ISFETs. *Biosensors and Bioelectronics*, 19(2) (2003), 131-135.

A novel urea biosensor based on immobilised recombinant urease as sensitive element and ion sensitive field effect transistor as transducer was developed. Recombinant urease from *E. coli* with an increased K_m was photoimmobilised in PVA/SbQ (poly(vinyl alcohol) containing styrylpyridinium) membrane and has demonstrated quite good performance as biosensitive element. Enzymatic field effect transistors based on such a bioselective element were studied in model buffer solutions. This biosensor demonstrated an extended dynamic range up to 80 mM, a quite good reproducibility (standard deviation of the sensor responses was approximately 2.5%, $n=20$ for urea concentration 10 mM) and a high stability. Such characteristics fit with the analytical requirements needed for urea control in plasma and liquids used during renal dialysis.

Anil K. Deisingh and Michael Thompson. Biosensors for the detection of bacteria. *Canadian Journal of Microbiology*, 50(2) (2004), 1-9.

This review will consider the role of biosensors towards the detection of infectious bacteria, although non-infectious ones will be considered where necessary. Recently, there has been a heightened interest in developing rapid and reliable methods of detection. This is especially true for detection of organisms involved in bioterrorism, food poisoning, and clinical problems such as antibiotic resistance. Biosensors can assist in achieving these goals, and sensors using several of the different types of transduction modes are discussed: electrochemical, high frequency (surface acoustic wave), and optical. The paper concludes with a discussion of three areas that may make a great impact in the next few years: integrated (lab-on-a-chip) systems, molecular beacons, and aptamers.

Caroline Michela, Fabienne Battaglia-Bruneta, Canh Tran Minhb, Mireille Bruschi and Ioannis Ignatiadis. (BRGM, Service Environnement Industriel et Procédés Innovants, Unité Biotechnologies, 3 Avenue Claude Guillemin, BP 6009, 45060, Orleans Cedex 02, France. Ecole Nationale Supérieure des Mines de St-Etienne, Centre SPIN, 158 Cours Fauriel, 42023, Saint Etienne Cedex 02, France. CNRS,

IBSM, Laboratoire de Bioénergétique et Ingénierie des Protéines, 31 chemin Joseph Aiguier, 13402, Marseille Cedex 20, France). Amperometric cytochrome c3-based biosensor for chromate determination. Biosensors and Bioelectronics, 19(4) (2003), 345-352.

The chromate reductase activity of cytochrome c3 (Cyt c3, Mr 13000), isolated from the sulfate-reducing bacterium *Desulfomicrobium norvegicum*, was used to develop an amperometric biosensor to measure chromate (CrO₄²⁻) bioavailability. The performance of various biosensor configurations for qualitative and quantitative determination of Cr(VI) was studied. Biosensor properties depend on the technique used to immobilize the enzyme on the electrode (glassy carbon electrode). Immobilization of Cyt c3 by entrapment in poly 3,4-ethylenedioxythiophene films denatured the enzyme, while application of an adsorption technique did not affect enzyme activity but the detection range was limited. The best results were obtained with dialysis membranes, which allowed the determination of Cr(VI) from 0.20 to 6.84 mg l⁻¹ (3.85–132 M) with a sensitivity of 35 nA mg⁻¹ l (1.82 nA M⁻¹). No interference was observed with As(V), As(III) and Fe(III). Only a small amount of Cyt c3 (372 ng of protein) was needed for this biosensor.

Daniel Filippini, Tony P. M. Anderssonb, Samuel P. S. Svenssonb, Ingemar Lundströma. (Division of Applied Physics, Institute of Physics and Measurement Technology, Linköping University, SE-581 83, Linköping, Sweden. Division of Pharmacology, Department of Medicine and Care, Faculty of Health Sciences, Linköping University, SE-581 85, Linköping, Sweden). Microplate based biosensing with a computer screen aided technique. Biosensors and Bioelectronics, 19(1) (2003), 9-19.

Melanophores, dark pigment cells from the frog *Xenopus laevis*, have the ability to change light absorbance upon stimulation by different biological agents. Hormone exposure (e.g. melatonin or -melanocyte stimulating hormone) has been used here as a reversible stimulus to test a new compact microplate reading platform. As an application, the detection of the asthma drug formoterol in blood plasma samples is demonstrated. The present system utilizes a computer screen as a (programmable) large area light source, and a standard web camera as recording media enabling even kinetic microplate reading with a versatile and broadly available platform, which suffices to evaluate numerous bioassays. Especially in the context of point of care testing or self testing applications these possibilities become advantageous compared with highly dedicated comparatively expensive commercial systems.

Graham H. Crossa, Andrew A. Reevesa, Stuart Branda, Jonathan F. Popplewellb, Louise L. Peelb, Marcus J. Swannb, Neville J. Freeman. (Department of Physics, University of Durham, South Road, Durham DH1 3LE, UK. Farfield Sensors Ltd., Salford University Business Park, Leslie Hough Way, Salford, Greater Manchester M6 6AJ, UK). A new quantitative optical biosensor for protein characterization. Biosensors and Bioelectronics, 19(4) (2003), 383-390.

A new optical biosensor is described based on a dual waveguide interferometric technique. By addressing the waveguide structure with alternate polarisations the optogeometrical properties (density and thickness) of adsorbed protein layers at the sensor (solid)-liquid interface have been determined. Differences in the waveguide mode dispersion between the transverse electric (TE) and transverse magnetic (TM) modes allow unique solutions for adlayer thickness and refractive index to be determined at all stages during the formation process. The technique has been verified using standard protein systems and by comparing the data with published work using X-ray crystallography and neutron reflection techniques. The data obtained was found to be in excellent agreement with previously reported X-ray experiments given that typical film thicknesses for streptavidin layers were in the range

5.5–6.5 nm compared with the short axis crystal structure of between 4.8 and 5.6 nm. The precision of the measurements taken was of the order of 40 pm with respect to adsorbed adlayer thicknesses. This biosensor approach provides measurements of both thickness and density of adlayers to a high precision, simultaneously and in real time enabling detail of the structure and function of proteins to be elucidated. From such data it is possible to obtain information on the orientation, distortion and efficiency of immobilisation procedures as well as the interaction event of interest. The technique is expected to find utility with those interested in protein structure and function. This is an area of growing importance within the life sciences as the demand for quantitative analytical techniques increases with the growth in 'proteomics'.

Jiantao Pang, Chunhai Fan, Xinjian Liu, Ting Chen, Genxi Li. (Department of Biochemistry and National Key Laboratory of Pharmaceutical Biotechnology, Nanjing University, Nanjing 210093, China). Nitric oxide biosensor based on the multi-assembly of hemoglobin/montmorillonite/polyvinyl alcohol at a pyrolytic graphite electrode. Biosensors and Bioelectronics, 19(5) (2003), 441-445.

Direct electron transfer of hemoglobin (Hb) can be achieved in a Hb/montmorillonite (MMT)/polyvinyl alcohol multi-assembly at a pyrolytic graphite (PG) electrode. Accordingly, a novel nitric oxide (NO) biosensor is proposed. The reduction of NO is observed at a potential of approximately -783 mV (vs. SCE) at pH 5.5. At optimum pH, this biosensor shows a wide linear range of 1.0×10^{-6} – 2.5×10^{-4} mol/l with a detection limit of 5.0×10^{-7} mol/l. The sensor-to-sensor reproducibility is good considering a relative standard deviation of 3.5% in five independent determinations at 5.0×10^{-5} mol/l NO. The modified electrode is conveniently constructed and durable in long-term operations.

Kwang-Soup Song, Toshikatsu Sakaia, b, Hirofumi Kanazawaa, Yuta Arakia, Hitoshi Umezawaa, Minoru Tachikia, Hiroshi Kawaradaa. (School of Science and Engineering, Waseda University, 3-4-1 Okubo, Shinjuku-ku, Tokyo 169-8555, Japan. CREST, Japan Science and Technology Corporation (JST), 5-3 Yonban, Chiyoda-ku, Tokyo 102-0081, Japan). Cl⁻ sensitive biosensor used electrolyte-solution-gate diamond FETs. Biosensors and Bioelectronics, 19(2) (2003), 137-140.

We have investigated the electrolyte-solution-gate field effect transistors (SGFETs) used hydrogen terminated (H-terminated) or partially oxygen terminated (O-terminated) polycrystalline diamond surface in the Cl⁻ and Br⁻ ionic solutions. The H-terminated channel SGFETs are insensitive to pH values in electrolyte solutions. The threshold voltages of the diamond SGFETs shift according to the density of Cl⁻ and Br⁻ ions about 30 mV/decade. One of the attractive biomedical applications for the Cl⁻ sensitive SGFETs is the detection of chloride density in blood or in sweat especially in the case of cystic fibrosis. The sensitivities of Cl⁻ and Br⁻ ions have been lost on the partially O-terminated diamond surface. These phenomena can be explained by the polarity of surface change on the H-terminated and the O-terminated surface.

M. Schwenk, T. Gabrio, O. Pöpke, T. Wallenhorst. (Landesgesundheitsamt Baden-Württemberg, Wiederholdstrasse 15, 70174 Stuttgart, Germany. Ergo Forschungsgesellschaft mbH, Geierstrasse 1, 22305 Hamburg, Germany). Human biomonitoring of polychlorinated biphenyls and polychlorinated dibenzodioxins and dibenzofuranes in teachers working in a PCB-contaminated school. Chemosphere, 47(2) (2002), 229-233.

Eighteen teachers from a highly contaminated school and 11 teachers from a control school participated in this study. Total polychlorinated biphenyl (PCB) indoor air concentration (six indicator congeners $\times 5$) was beyond 12 000 ng/m³ in some rooms of the contaminated

school. PCB 28 and PCB 52 were the prevailing congeners. Whole blood was taken from each participant by the local health authority, pooled in two groups and analysed for the six PCB indicator congeners, non-ortho PCBs and polychlorinated dibenzodioxins (PCDD)/polychlorinated dibenzofurans (PCDF). Blood analysis showed elevated mean PCB 28 and PCB 52 levels for the exposed group, however the two groups were almost identical with regard to PCB 101, 138, 153 and 180. Moreover no difference can be observed for the concentration of non-ortho PCBs and PCDD/PCDF. The data support the finding, that heavy indoor air contamination with low chlorinated PCBs causes an increase of PCB 28 and PCB 52 blood levels. However, this increment was small compared to their total PCB load.

P. C. J. Roach, D. K. Ramsdena, J. Hughesb, P. Williamsa. (Biotechnology Centre, School of Applied Sciences, University of Huddersfield, Queensgate, Huddersfield, West Yorkshire, HD1 3DH, UK. Ciba Specialty Chemicals, P.O. Box 38, Low Moor, Bradford, West Yorkshire, BD12 0JZ, UK). Development of a conductimetric biosensor using immobilised *Rhodococcus ruber* whole cells for the detection and quantification of acrylonitrile. *Biosensors and Bioelectronics*, 19(1) (2003), 73-78.

A conductimetric biosensor for the detection of acrylonitrile in solution was designed and characterised using whole cells of *Rhodococcus ruber* NCIMB 40757, which were immobilised into a disc of dimethyl silicone sponge (ImmobaSil™). The biosensor described was capable of the detection and quantification of acrylonitrile in aqueous solution, having a linear response to concentrations between 2 and 50 mM (106–2650 ppm) acrylonitrile. The biosensor has been shown to be reproducible with respect to the data obtained over a number of days, and retains stability for a minimum period of at least 5 days before recalibration of the biosensor is required.

Seiji Shibasaki, Atsuo Tanakab, Mitsuyoshi Ueda. (Department of Applied Chemistry, Kobe City College of Technology, Gakuen-higashimachi, Nishi-ku, Kobe 651-2194, Japan. Department of Synthetic Chemistry and Biological Chemistry, Graduate School of Engineering, Kyoto University, Yoshida, Sakyo-ku, Kyoto 606-8501, Japan). Development of combinatorial bioengineering using yeast cell surface display--order-made design of cell and protein for bio-monitoring. *Biosensors and Bioelectronics*, 19(2) (2003), 123-130.

A genetic system to display proteins as their active and functional forms on the cell surface of yeast, *Saccharomyces cerevisiae*, has been exploited. Surface-engineered (arming) cells displaying amylase or cellulase could assimilate starch or cellulose as the sole carbon source, although *S. cerevisiae* can not intrinsically assimilate them. Arming cells with a green fluorescent protein (GFP) from *Aequorea victoria* can emit green fluorescence from the cell surface in response to the environmental conditions. From these results, we attempted to construct a system to monitor the foreign protein production in yeast by simultaneous displaying the enhanced GFP (EGFP). The expression in yeast of the *Escherichia coli* -galactosidase-encoding gene was examined as an example of intracellular production and that of the human interferon- (ω , IFN-)-encoding gene as an example of extracellular production. Their productions and the simultaneous surface-display of EGFP as a reporter were controlled by the same promoter, GAL1. The relationship among fluorescence signals and their productions was evaluated. The surface-display system, unlike one using tag-proteins, would be able to facilitate the monitoring of native protein productions in bioprocesses using living cells in real time by the combination of promoters and GFP variants.

Yang Liu, Xiao Yu, Rui Zhao, Di-Hua Shangguan, Zuyi Bo, Guoquan Liu. (Center for Molecular Science, Institute of Chemistry, Chinese Academy of Sciences, Beijing 100080, China). Quartz crystal biosensor for real-time monitoring of molecular

recognition between protein and small molecular medicinal agents. *Biomass and Bioenergy*, 25(6) (2003), 623-636.

A quartz crystal microbalance (QCM) biosensor integrated into a flow injection analysis (FIA) system was used for the real-time investigation of molecular recognition between a protein and small molecular medicinal agents. Two sulfa-drugs, sulfamethazine (SMZ) and sulfamethoxazole (SMO), were, respectively, immobilized on the gold electrodes of the piezoelectric crystals using appropriate procedures based on self-assembly of the dithiothreitol (DTT). The binding interactions of the two immobilized drug ligands, with various proteins in solution, were followed as changes in the resonant frequency of the modified crystals. Results obtained from this rapid screen analysis clearly indicated that the two drug ligands appeared quite different in this molecular recognition procedure although their structures were similar. SMZ-immobilized sensor showed specific interaction only with IgG, while SMO-immobilized sensor showed negligible specific binding with IgG, but binding with trypsin and chymotrypsin. Further studies on the specific interaction between immobilized SMZ and three different species of IgG—human IgG, goat IgG and mouse IgG were carried out and the marked species-dependent difference was observed. The resultant sensorgrams were rapidly analyzed by using an in-house kinetic analysis software based on genetic algorithm (GA) to derive both the kinetic rate constants (k_{ass} and k_{diss}) and equilibrium association constants (K_A) for IgG–SMZ interactions. For the interactions, K_A were 5.48×10^5 , 2.75×10^5 and 1.86×10^5 M⁻¹ for human IgG, goat IgG and mouse IgG, respectively. The kinetic data provided further insight into the structural/functional relationships of different IgG on a molecular level.

Yuh-Ming Uanga, Tse-Chuan Chou. (Chemical Engineering Department, Wu-Feng Institute of Technology, Chia-Yi, Taiwan, ROC. Chemical Engineering Department, National Cheng Kung University, Tainan 701, Taiwan, ROC). Fabrication of glucose oxidase/polypyrrole biosensor by galvanostatic method in various pH aqueous solutions. *Biosensors and Bioelectronics*, 19(3) (2003), 141-147.

The pH effect of pyrrole electropolymerization in the presence of glucose oxidase (GODx) on the performance and characteristic of galvanostatically fabricated glucose oxidase/polypyrrole (Ppy) biosensor is reported. Preparing the GODx/Ppy biosensors in 0.1 M KCl saline solution with various pH containing 0.05 M pyrrole monomer and 0.5 mg/ml GODx at 382 A/cm² current density for 100 mC/cm² film thickness, both the galvanostatic responses and characteristics of these resulted biosensors were obtained. The results revealed that the galvanostatic glucose biosensor fabricated at neutral pH condition exhibited much higher sensitivity than those fabricated at lower or higher pH conditions, and had a good linearity form zero to 10 mM glucose with the sensitivity of 7 nA/mM. Finally, the long-term stability and the kinetic parameters, Michaelis constant and maximum current, of this biosensor were also reported.

Biotechnology Policy Issue

A. P. Mitra, C. Sharma. (National Physical Laboratory, Dr. K.S. Krishnan Road, New Delhi-110012, India). Indian aerosols: present status. *Chemosphere*, 49(9) (2002), 1175-1190.

This article presents the status of aerosols in India based on the research activities undertaken during last few decades in this region. Programs, like International Geophysical Year (IGY), Monsoon Experiment (MONEX), Indian Middle Atmospheric Program (IMAP) and recently conducted Indian Ocean Experiment (INDOEX), have thrown new lights on the role of aerosols in global change. INDOEX has proved that the effects of aerosols are no longer confined to the local levels but extend at regional as well as global scales due to occurrence

of long range transportation of aerosols from source regions along with wind trajectories. The loading of aerosols in the atmosphere is on rising due to energy intensive activities for developmental processes and other anthropogenic activities. One of the significant observation of INDOEX is the presence of high concentrations of carbonaceous aerosols in the near persistent winter time haze layer over tropical Indian Ocean which have probably been emitted from the burning of fossil-fuels and biofuels in the source region. These have significant bearing on the radiative forcing in the region and, therefore, have potential to alter monsoon and hydrological cycles. In general, the SPM concentrations have been found to be on higher sides in ambient atmosphere in many Indian cities but the NO_x concentrations have been found to be on lower side. Even in the haze layer over Indian Ocean and surrounding areas, the NO_x concentrations have been reported to be low which is not conducive of O₃ formation in the haze/smog layer. The acid rain problem does not seem to exist at the moment in India because of the presence of neutralizing soil dust in the atmosphere. But the high particulate concentrations in most of the cities' atmosphere in India are of concern as it can cause deteriorated health conditions.

Ingle ST, Pachpande B G, Mali D S. (School of Environmental and Earth Sciences, North Maharashtra University Jalgaon-425 001, India). Impact of Ganpati festival on ambient noise levels of Kolhapur city. Indian Journal of Environmental Protection, 22(3) (2002), 308-314.

Ambient noise level monitoring was carried out at various locations of the city during Ganpati festival of 2000. The obtained was used to compute various noise parameters, namely: equivalent continuous levels (Leq); percentile no levels (L10, L50 and L90); NC (noise climate) and noise pollution level (Lnp). A great variation in the noise climate observed at various locations of the city. The comparison data shows that the noise levels at various locations of the city are more than the prescribed limit for residential area. Very high noise levels were recorded on the first and last day of festival. At all the sampling locations higher noise levels were observed in the evening during the study period. The study urges the need for awareness about health hazard of noise pollution.

Lal B, Mishra S, Bhattacharya D, Sarma P M. Biotechnological approach to manage oily sludge. In Proceedings of the 4th International Petroleum Conference and Exhibition, (2001), 280.

An indigenous bacterial consortium was developed by combining the cultures of five bacterial strains, which could biodegrade crude oil and oily sludge. This indigenous bacterial consortium was designated as 'Oilzapper'. Crude oil and oily sludge degrading efficiency of Oilzapper was tested under laboratory and field conditions. A full-scale study on in situ bioremediation of oily sludge was conducted at an oil refinery. The indigenous population of hydrocarbon degrading bacteria in soil at full scale bioremediation site was only 1000-10000 cfu/g soil. Treatment plots A1, A2 and A3 of block A were treated with Oilzapper and nutrients, resulted in 96.2%, 90.6% and 90.3% biodegradation of TPH respectively in one year as compared to only 14.3% biodegradation of TPH in control block. Similarly the plots B1, B2 and B3 of block B were also treated with Oilzapper and nutrients, which revealed 90.6%, 89.6% and 87.8% biodegradation of TPH in soil in one year. The population of *Acinetobacter baumannii* strains (constituent of Oilzapper) was stable in soil at bioremediation site even after one year of its application. Physical and chemical properties of soil of bioremediation site improved significantly in one year. A similar study on bioremediation of oily sludge with application of Oilzapper was also conducted at various oil refineries and the results are highly encouraging.

Mohindru V K, Singh Kawaljeet, Jena B K, Siddiqui N A, Kulkarni A G. (Central Pulp and Paper Research Institute, Saharanpur- 247 001, India). Comparison of 5-year micro-meteorological data over a paper industry and its role in environmental

quality representation. *Indian Journal of Environmental Protection*, 22(3) (2002), 277-283.

Micro-meteorological parameters are one of the important data: which is collected and analysed for its effect on pollutants emitted by the industrial activity. However, there is little knowledge in the paper industry, regarding the interpretation of such data from the point of view of environmental impact assessment. The present paper attempts to put forward a comparison of meteorological data collected at title site of a large paper mill. The data was collected over a period of five years for the worst months of winter (November to February, between 1996-2000). The data is presented in the form of windrose diagrams and charts to bring out the salient variation of meteorological conditions over the test area and its implication in interpreting the environmental impact of various us pollutants being generated from the mill. The study is presented in a manner so as to give the executive of the industry an insight into the importance of micro— meteorological studies conducted at Site. Micro-meteorological data go to indicate that a persistent high value of calm period is observed for November-December. During these two months, the ESPs attached to all boilers must be operated at peak power levels. In fact, it would be best to plan the annual maintenance of all ESPs in September-October so as to obtain peak performance in December.

Sinha S and Sreekesh S. (TERI, Habitat Place, Lodhi Road, New Delhi-110003, India). Air quality status and management options for the mining belt of Goa. *Indian Journal of Environmental Protection*, 22(3) (2002), 241-253.

Mine operations, overburdened dumps, dry screening, and transportation of fine ore are the major sources of particulate matter in the mining belt of Goa. In mines, loading, unloading, drilling, blasting, and truck/dumper movements on haul roads are the main sources of dust. SPM (suspended particulate matter) levels are poor, and exceeding standards in 20 villages. SPM levels are also exceeding the concentration of 500 µg/m³ in almost all operating mines. Operator's exposure to dust levels is reported to be varying from 0.87 to 5.68 mg/ m³. The drill and chute operators are exposed to dust levels higher than the standard level. SPM concentration along the road corridors is estimated and it exceeds the ambient air quality standards for residential areas. AQI (air quality index,) estimated for 27 villages shows poor and bad AQI. In order to improve the air quality in the mining belt, in addition to strengthening the existing control measures adopted by the mining companies, additional measures need to be undertaken to control dust generation and its dispersion. These measures are: green belts around all mines experimentation with chemical spraying in permanent haul roads, improved mine and public roads, and vegetation pf dumps in a scientific manner.

U. F. Wehmeier, W. Piepersberg. (Chemische Mikrobiologie, Bergische Universitaet Wuppertal, Gauß-Straße 20, 42097 Wuppertal, Germany). Biotechnology and molecular biology of the α-glucosidase inhibitor acarbose. *Applied Microbiology and Biotechnology*, 63(6) (2004), 613 – 625.

The α-glucosidase inhibitor acarbose, O-{4,6-dideoxy-4[1s-(1,4,6/5)-4,5,6-trihydroxy-3-hydroxymethyl-2-cyclohexen-1-yl]-amino-α-d-glucopyranosyl}-(1->4)-O-α-d-glucopyranosyl-(1->4)-d-glucopyranose, is produced in large-scale fermentation by the use of strains derived from *Actinoplanes* sp. SE50. It has been used since 1990 in many countries in the therapy of diabetes type II, in order to enable patients to better control blood sugar contents while living with starch-containing diets. Thus, it is one of the latest successful products of bacterial secondary metabolism to be introduced into the pharmaceutical world market. Cultures of *Actinoplanes* sp. also produce various other acarbose-like components, of which component C is hard to separate during downstream processing, which is one of the most modern work-up processes developed to date. The

physiology, genetics and enzymology of acarbose biosynthesis and metabolism in the producer have been studied to some extent, leading to the proposal of a new pathway and metabolic cycle, the "carbophore". These data could give clues for further biotechnological developments, such as the suppression of side-products, enzymological or biocombinatorial production of new metabolites and the engineering of production rates via genetic regulation in future.

Biotransformation

Bharat Bhushan, Louise Paquet, Jim C. Spain, Jalal Hawari. (Biotechnology Research Institute, National Research Council of Canada, Montreal, Quebec H4P 2R2, Canada, U.S. Air Force Research Laboratory, Tyndall Air Force Base, Florida 32403). Biotransformation of 2,4,6,8,10,12-Hexanitro-2,4,6,8,10,12-Hexaazaisowurtzitane (CL-20) by Denitrifying *Pseudomonas* sp. Strain FA1. *Applied and Environmental Microbiology*, 69(10) (2003), 337-6339.

The microbial and enzymatic degradation of a new energetic compound, 2,4,6,8,10,12-hexanitro-2,4,6,8,10,12-hexaazaisowurtzitane (CL-20), is not well understood. Fundamental knowledge about the mechanism of microbial degradation of CL-20 is essential to allow the prediction of its fate in the environment. In the present study, a CL-20-degrading denitrifying strain capable of utilizing CL-20 as the sole nitrogen source, *Pseudomonas* sp. strain FA1, was isolated from a garden soil. Studies with intact cells showed that aerobic conditions were required for bacterial growth and that anaerobic conditions enhanced CL-20 biotransformation. An enzyme(s) involved in the initial biotransformation of CL-20 was shown to be membrane associated and NADH dependent, and its expression was up-regulated about 2.2-fold in CL-20-induced cells. The rates of CL-20 biotransformation by the resting cells and the membrane-enzyme preparation were 3.2 ± 0.1 nmol h⁻¹ mg of cell biomass⁻¹ and 11.5 ± 0.4 nmol h⁻¹ mg of protein⁻¹, respectively, under anaerobic conditions. In the membrane-enzyme-catalyzed reactions, 2.3 nitrite ions (NO₂⁻), 1.5 molecules of nitrous oxide (N₂O), and 1.7 molecules of formic acid (HCOOH) were produced per reacted CL-20 molecule. The membrane-enzyme preparation reduced nitrite to nitrous oxide under anaerobic conditions. A comparative study of native enzymes, deflavoenzymes, and a reconstituted enzyme(s) and their subsequent inhibition by diphenyliodonium revealed that biotransformation of CL-20 is catalyzed by a membrane-associated flavoenzyme. The latter catalyzed an oxygen-sensitive one-electron transfer reaction that caused initial N denitration of CL-20.

Bharat Bhushan, Sandra Trott, Jim C. Spain, Annamaria Halasz, Louise Paquet, Jalal Hawari. (Biotechnology Research Institute, National Research Council of Canada, Montreal, Quebec H4P 2R2, Canada, 1 U.S. Air Force Research Laboratory, Tyndall Air Force Base, Florida 32403). Biotransformation of Hexahydro-1,3,5-Trinitro-1,3,5-Triazine (RDX) by a Rabbit Liver Cytochrome P450: Insight into the Mechanism of RDX Biodegradation by *Rhodococcus* sp. Strain DN22. *Applied and Environmental Microbiology*, 69(3) (2003), 1347-1351.

A unique metabolite with a molecular mass of 119 Da (C₂H₅N₃O₃) accumulated during biotransformation of hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX) by *Rhodococcus* sp. strain DN22 (D. Fournier, A. Halasz, J. C. Spain, P. Fiurasek, and J. Hawari, *Appl. Environ. Microbiol.* 68:166-172, 2002). The structure of the molecule and the reactions that led to its synthesis were not known. In the present study, we produced and purified the unknown metabolite by biotransformation of RDX with *Rhodococcus* sp. strain DN22 and identified the molecule as 4-nitro-2,4-diazabutanal using nuclear magnetic resonance and elemental analyses. Furthermore, we tested the hypothesis that a cytochrome P450 enzyme was

responsible for RDX biotransformation by strain DN22. A cytochrome P450 2B4 from rabbit liver catalyzed a very similar biotransformation of RDX to 4-nitro-2,4-diazabutanal. Both the cytochrome P450 2B4 and intact cells of *Rhodococcus* sp. strain DN22 catalyzed the release of two nitrite ions from each reacted RDX molecule. A comparative study of cytochrome P450 2B4 and *Rhodococcus* sp. strain DN22 revealed substantial similarities in the product distribution and inhibition by cytochrome P450 inhibitors. The experimental evidence led us to propose that cytochrome P450 2B4 can catalyze two single electron transfers to RDX, thereby causing double denitration, which leads to spontaneous hydrolytic ring cleavage and decomposition to produce 4-nitro-2, 4-diazabutanal. Our results provide strong evidence that a cytochrome P450 enzyme is the key enzyme responsible for RDX biotransformation by *Rhodococcus* sp. strain DN22.

Byung-Taek Oh, Patrick J. Shea, Rhae A. Drijber, Galina K. Vasilyeva, Gautam Sarath. (School of Chemical Engineering, BK21, Seoul National University, San 56-1, Shinlim-dong, Gwanak-gu, Seoul 151-744, Korea. School of Natural Resources, BK21, Seoul National University, San 56-1, Shinlim-dong, Gwanak-gu, Seoul 151-744, Korea. Department of Agronomy and Horticulture, University of Nebraska, Lincoln, NE 68583-0915, USA. Institute of Physicochemical and Biological Problems in Soil Science, Russian Academy of Sciences, Pushchino, Russia 142292. USDA-ARS & Department of Biochemistry, University of Nebraska, Lincoln, NE 68583-0664, USA). TNT Biotransformation and Detoxification by a *Pseudomonas aeruginosa* Strain. *Biodegradation*, 14(5) (2003), 309-319.

Successful microbial-mediated remediation requires transformation pathways that maximize metabolism and minimize the accumulation of toxic products. *Pseudomonas aeruginosa* strain MX, isolated from munitions-contaminated soil, degraded 100 mg TNT L⁻¹ in culture medium within 10 h under aerobic conditions. The major TNT products were 2-amino-4,6-dinitrotoluene (2ADNT, primarily in the supernatant) and 2,2'-azoxytoluene (2,2'AZT, primarily in the cell fraction), which accumulated as major products via the intermediate 2-hydroxylamino-4,6-dinitrotoluene (2HADNT). The 2HADNT and 2,2'AZT were relatively less toxic to the strain than TNT and 2ADNT. Aminodinitrotoluene (ADNT) production increased when yeast extract was added to the medium. While TNT transformation rate was not affected by pH, more HADNTs accumulated at pH 5.0 than at pH 8.0 and AZTs did not accumulate at the lower pH. The appearance of 2,6-diamino-4-nitrotoluene (2,6DANT) and 2,4-diamino-6-nitrotoluene (2,4DANT); dinitrotoluene (DNT) and nitrotoluene (NT); and 3,5-dinitroaniline (3,5DNA) indicated various routes of TNT metabolism and detoxification by *P. aeruginosa* strain MX.

C. M. Kao, J. K. Liu, H. R. Lou, C. S. Lin, S. C. Chen. (Institute of Environmental Engineering, National Sun Yat-Sen University, Kaohsiung, Taiwan, ROC. Department of Biological Science, National Sun Yat-Sen University, Kaohsiung, Taiwan, ROC. Department of Medical Technology, Fooyin University, Kaohsiung, Taiwan, ROC. Department of Marine Resources, National Sun Yat-Sen University, Kaohsiung, Taiwan, ROC. Department of Medicine, China Medical College, 91 Huseh-Shih Road, Taichung 400, Taiwan, ROC). Biotransformation of cyanide to methane and ammonia by *Klebsiella oxytoca*. *Chemosphere*, 50(8) (2003), 1055-1061.

Klebsiella oxytoca, isolated from cyanide-containing industrial wastewater, was shown to be able to biodegrade cyanide to non-toxic endproducts using cyanide as the sole nitrogen source. In this study, ammonia was one of the detected endproduct of cyanide biodegradation by the concentrated resting cells of *K. oxytoca*. Moreover, cyanide has been shown to be biotransformed to methane through the actions of concentrated resting cells. Biodegradation of cyanide by cell-free extracts was not observed, which might be due to the

inactivation of nitrogenase (an oxygen-labile enzyme) caused by the oxygen exposure after cell disruption. Results show that the cyanide consumption by resting cells of *K. oxytoca* was induced when the pretreatment of these cells with cyanide was conducted. However, the cyanide-degrading capability of resting cells pretreated with ammonia was inhibited. The inhibition of cyanide degradation by resting cells of *K. oxytoca* was affected by the ammonia concentration. This might result from the suppression of nitrogenase activity of *K. oxytoca* by ammonia since nitrogenase was suggested to be the sole cyanide-degrading enzyme during the cyanide degradation process. Results from this study also show that the processes of cyanide biodegradation and ammonia production by resting cells occurred simultaneously. This suggests that the utilization of cyanide as nitrogen source by *K. oxytoca* might proceed using ammonia as an assimilatory substrate.

Céline Tixier, Martine Sancelme, Selim Aït-Aïssa, Pascale Widehem, Frédérique Bonnemoy, Annie Cuer, Nicole Truffaut, Henri Veschambre. (Synthèse et Etude de Systèmes à Intérêt Biologique, UMR 6504, Université Blaise Pascal, 63177 Aubière Cedex, France. Laboratoire de Biologie des Protistes, UPRES A 6023, Université Blaise Pascal, 63177 Aubière Cedex, France. Laboratoire de Génétique Microbienne, UMR 6022, Université de Technologie de Compiègne, BP 20529, 60205 Compiègne Cedex, France). Biotransformation of phenylurea herbicides by a soil bacterial strain, *Arthrobacter* sp. N2: structure, ecotoxicity and fate of diuron metabolite with soil fungi. *Chemosphere*, 46(4) (2002), 519-526.

In order to assess the influence of the aromatic substitution on the ability of a soil bacterial strain, *Arthrobacter* sp. N2, to degrade phenylurea herbicides, biotransformation assays were performed in mineral medium with resting cells of this soil bacterial strain on three phenylurea herbicides (diuron, chlorotoluron and isoproturon). Each herbicide considered, led to the formation of only one metabolite detected by HPLC analysis. After isolation, the metabolites were identified by NMR and MS, as the corresponding substituted anilines. According to the Microtox[®] test (realized on the bacterium *Vibrio fischeri*), these metabolites presented non-target toxicity far more important (up to 600 times higher for 4-isopropylaniline) than the parent molecule. For isoproturon and chlorotoluron, the amount of substituted anilines obtained at the end of the biotransformation was very low, whereas the biotransformation of diuron into 3,4-dichloroaniline was almost quantitative. In this last case, the degradation product accumulated in the medium. In soil, other microorganisms are present that might degrade it. So the biotransformation of 3,4-dichloroaniline was then tested with four fungal strains: *Aspergillus niger*, *Beauveria bassiana*, *Cunninghamella echinulata* var. *elegans* and *Mortierella isabellina*. The aniline was further transformed with all the microorganisms tested. Only one metabolite was detected by HPLC analysis and after isolation, it was identified to be 3,4-dichloroacetanilide. This acetylated compound led to biological effects less important on *V. fischeri* than 3,4-dichloroaniline. These results stress the importance of identifying the degradation products to assess the impact of a polluting agent. Indeed, the pollutant may undergo transformation yielding compounds more toxic than the parent molecule.

D.Y. Bojinova, R.G. Velkova. (University of Chemical Technology and Metallurgy, 8 Kliment Ohridski Blvd, 1756 Sofia, Bulgaria). Bioleaching of Metals from Mineral Waste Product. *Acta Biotechnologica*, 21(3) (2001), 275 – 282.

The possibility of bioleaching Al, K, Na, Ca and Mg using microorganisms of the *Thiobacillus* thiooxidans group from industrial waste product (IWP) of copper ore flotation from the company MEDET was studied. The aim of the investigations was to establish the possible application of a combined method for processing IWP. The preliminary mechanical activation in combination with bioleaching resulted in a high extent of extraction of useful components. It was established that the removal of useful components from mechanically activated IWP

is improved compared to non-activated IWP. The effect of the concentration of Al-containing waste product, of incubation time and time of preliminary mechanical activation on the extraction degree (α) [% w/w] of useful elements was investigated. The maximum degree of extraction of Al was achieved on Day 28 and its value reached 71% for industrial waste product mechanically activated for 4 hours. The maximum degrees of extraction of K and Na in the case of industrial waste product mechanically activated for 4 hours were achieved on Day 7 of the incubation period and their values were 78% and 91%, respectively. Under the conditions of bioleaching only Si had a low degree of extraction, accounting for 2.5%. The ability of microorganisms to leach aluminium could be used for the extraction of metals from nonbauxite raw materials and Al-containing waste product not treatable by means of the BAYER method.

Dai Kitamoto, Toru Ikegami, Gaby Tiemi Suzuki, Akira Sasaki, Yu-ichiro Takeyama, Yasushi Idemoto, Nobuyuki Koura, Hiroshi Yanagishita. (National Institute of Advanced Industrial Science and Technology, Central 5, Higashi 1-1, Tsukuba, Ibaraki 305-8565, Japan. Department of Industrial Chemistry, Science University of Tokyo, 2641 Yamazaki, Noda, Chiba 278-8510, Japan). Microbial conversion of n-alkanes into glycolipid biosurfactants, mannosylerythritol lipids, by *Pseudozyma (Candida antarctica)*. *Biotechnology Letters*, 23(20) (2001), 1709-1714.

n-Alkanes ranging from C12 to C18 were converted into glycolipid biosurfactants, mannosylerythritol lipids (MEL), by resting cells of *Pseudozyma (Candida) antarctica* T-34. The highest yield (0.87 g g⁻¹ substrate) was obtained from 6% (v/v) of n-octadecane after 7 days reaction. The amount of MEL reached 140 g l⁻¹ by intermittent feeding of the substrate.

David J. Hopper, Lisa Cottrell. (Institute of Biological Sciences, University of Wales, Aberystwyth, Ceredigion SY23 3DD, United Kingdom). Alkylphenol Biotransformations Catalyzed by 4-Ethylphenol Methylhydroxylase. *Applied and Environmental Microbiology*, 69(6) (2003), 3650-3652.

4-Ethylphenol methylhydroxylase from *Pseudomonas putida* JD1 acts by dehydrogenation of its substrate to give a quinone methide, which is then hydrated to an alcohol. It was shown to be active with a range of 4-alkylphenols as substrates. 4-n-Propylphenol, 4-n-butylphenol, chavicol, and 4-hydroxydiphenylmethane were hydroxylated on the methylene group next to the benzene ring and produced the corresponding chiral alcohol as the major product. The alcohols 1-(4'-hydroxyphenyl)propanol and 1-(4'-hydroxyphenyl)-2-propen-1-ol, produced by the biotransformation of 4-n-propylphenol and chavicol, respectively, were shown to be R(+) enantiomers. 5-Indanol, 6-hydroxytetralin, 4-isopropylphenol, and cyclohexylphenol, with cyclic or branched alkyl groups, gave the corresponding vinyl compounds as their major products.

Henry H. Tabak, Richard Scharp, John Burckle, Fred K. Kawahara, Rakesh Govind. (U.S. Environmental Protection Agency, ORD, NRMRL, Cincinnati, OH 45268, USA. University of Cincinnati, Department of Chemical Engineering, Cincinnati, OH 45221, USA). Advances in biotreatment of acid mine drainage and biorecovery of metals: 1. Metal precipitation for recovery and recycle. *Biodegradation*, 14(6) (2003), 423-436.

Acid mine drainage (AMD), an acidic metal-bearing wastewater, poses a severe pollution problem attributed to post mining activities. The metals usually encountered in AMD and considered of concern for risk assessment are arsenic, cadmium, iron, lead, manganese, zinc, copper and sulfate. The pollution generated by abandoned mining activities in the area of Butte, Montana has resulted in the designation of the Silver Bow Creek-Butte Area as the largest Superfund (National Priorities List) site in the U.S. This paper reports the results of

bench-scale studies conducted to develop a resource recovery based remediation process for the clean up of the Berkeley Pit. The process utilizes selective, sequential precipitation (SSP) of metals as hydroxides and sulfides, such as copper, zinc, aluminum, iron and manganese, from the Berkeley Pit AMD for their removal from the water in a form suitable for additional processing into marketable precipitates and pigments. The metal biorecovery and recycle process is based on complete separation of the biological sulfate reduction step and the metal precipitation step. Hydrogen sulfide produced in the SRB bioreactor systems is used in the precipitation step to form insoluble metal sulfides. The average metal recoveries using the SSP process were as follows: aluminum (as hydroxide) 99.8%, cadmium (as sulfide) 99.7%, cobalt (as sulfide) 99.1% copper (as sulfide) 99.8%, ferrous iron (sulfide) 97.1%, manganese (as sulfide) 87.4%, nickel (as sulfide) 47.8%, and zinc (as sulfide) 100%. The average precipitate purity for metals, copper sulfide, ferric hydroxide, zinc sulfide, aluminum hydroxide and manganese sulfide were: 92.4, 81.5, 97.8, 95.6, 92.1 and 75.0%, respectively. The final produced water contained only calcium and magnesium and both sulfate and sulfide concentrations were below usable water limits. Water quality of this agriculturally usable water met the EPA's gold standard criterion.

Inge S. Fomsgaard, Anne G. Mortensen, Sandra C. K. Carlsen. (Department of Crop Protection, Research Centre Flakkebjerg, Danish Institute of Agricultural Sciences, Slagelse DK-4200, Denmark). Microbial transformation products of benzoxazolinone and benzoxazinone allelochemicals--a review. Chemosphere, 54(8) (2004), 1025-1038.

Cyclic hydroxamic acids and lactams are allelochemicals present in the common agricultural crops wheat, rye, and maize. The hydroxamic acids are mainly present in the plants as glucosides. Upon injury or insect attack or when exuded to the soil environment, the hydroxamic acids occur in their unstable agluconic form. In the first step in the transformation of hydroxamic acids, benzoxazolinones are formed spontaneously. It is necessary to elucidate the further microbial transformation of these compounds in the soil environment for a purposeful exploitation of the allelopathic properties of wheat, rye, and maize. In the present paper, the existing knowledge on microbial transformation products of benzoxazolin-2-one (BOA), 6-methoxy-benzoxazolin-2-one (MBOA), and 2-hydroxy-1,4-benzoxazin-3-one (HBOA) was reviewed. Three main groups of transformation products were identified: aminophenoxazinones, malonamic acids, and acetamides. Future research needs concerning the transformation of these chemicals in soil are discussed, when their properties for suppressing weeds and soil-borne diseases are going to be exploited.

Jörg Overhage, Alexander Steinbüchel, Horst Priefert. (Institut für Molekulare Mikrobiologie und Biotechnologie der Westfälischen Wilhelms-Universität Münster, D-48149 Münster, Germany). Highly Efficient Biotransformation of Eugenol to Ferulic Acid and Further Conversion to Vanillin in Recombinant Strains of *Escherichia coli*. Applied and Environmental Microbiology, 69(11) (2003), 6442-6446.

The *vaoA* gene from *Penicillium simplicissimum* CBS 170.90, encoding vanillyl alcohol oxidase, which also catalyzes the conversion of eugenol to coniferyl alcohol, was expressed in *Escherichia coli* XL1-Blue under the control of the *lac* promoter, together with the genes *calA* and *calB*, encoding coniferyl alcohol dehydrogenase and coniferyl aldehyde dehydrogenase of *Pseudomonas* sp. strain HR199, respectively. Resting cells of the corresponding recombinant strain *E. coli* XL1-Blue(pSKvaomPcalAmcalB) converted eugenol to ferulic acid with a molar yield of 91% within 15 h on a 50-ml scale, reaching a ferulic acid concentration of 8.6 g liter⁻¹. This biotransformation was scaled up to a 30-liter fermentation volume. The maximum production rate for ferulic acid at that scale was 14.4 mmol per h per liter of culture. The maximum concentration of ferulic acid obtained was 14.7 g liter⁻¹ after a

total fermentation time of 30 h, which corresponded to a molar yield of 93.3% with respect to the added amount of eugenol. In a two-step biotransformation, *E. coli* XL1-Blue(pSKvaomPcalAmcalB) was used to produce ferulic acid from eugenol and, subsequently, *E. coli*(pSKecheE/Hfcs) was used to convert ferulic acid to vanillin (J. Overhage, H. Priefert, and A. Steinbüchel, *Appl. Environ. Microbiol.* 65:4837-4847, 1999). This process led to 0.3 g of vanillin liter⁻¹, besides 0.1 g of vanillyl alcohol and 4.6 g of ferulic acid liter⁻¹. The genes *ehyAB*, encoding eugenol hydroxylase of *Pseudomonas* sp. strain HR199, and *azu*, encoding the potential physiological electron acceptor of this enzyme, were shown to be unsuitable for establishing eugenol bioconversion in *E. coli* XL1-Blue.

Kamlesh Kanwar, Anju Bhardwaj, Sandhya Agarwal & D R Sharma. Genetic transformation of *Robinia pseudoacacia* by *Agrobacterium tumefaciens*. *Indian Journal of Experimental Biology*, 41 (2003), 149-153.

Transgenic *Robinia pseudoacacia* plants were obtained by *Agrobacterium tumefaciens* mediated gene transfer. *Agrobacterium* strain LBA4404 harbouring a binary vector that contained the chimeric neomycin phosphotransferase II (NPTII) and b-glucuronidase (GUS) genes was co-cultivated with hypocotyl segments of in vitro raised seedlings of *Robinia*. Parameters important for high efficiency regeneration and transformation rates included type of explant, pre-conditioning of explants and appropriate length of co-cultivation period with *Agrobacterium*. A transformation frequency 16.67% was obtained by 48 hr of pre-conditioning followed by 48 hr of co-cultivation. Transformed tissue was selected by the ability to grow on kanamycin containing medium. Successful regeneration was followed after histochemical GUS assay for the detection of transgenic tissue. This transformation procedure has the potential to expand the range of genetic variation in *Robinia*.

Keishiro Yoshida, Kazuo Furihata, Hiroshi Habe, Hisakazu Yamane, Toshio Omori. (Central Laboratory, Lotte Co., Ltd., 3-1-1 Numakage, Urawa, Saitama 336-0027, Japan. Department of Applied Biological Chemistry, The University of Tokyo, 1-1-1 Yayoi, Bunkyo-ku, Tokyo 113-8657, Japan. Biotechnology Research Center, The University of Tokyo, 1-1-1 Yayoi, Bunkyo-ku, Tokyo 113-8657, Japan). Microbial transformation of 18 β -glycyrrhetic acid by *Sphingomonas paucimobilis* strain G5. *Biotechnology Letters*, 23(19) (2001), 1619-1624.

The effects of the oxygenase inhibitors, 1-aminobenzotriazole (ABT), ketoconazole, metyrapone and proadifen, on the metabolism of 18 β -glycyrrhetic acid (18 β -GRA) in *Sphingomonas paucimobilis* strain G5 were investigated. Strain G5 transformed 18 β -GRA into a major new metabolite (M-D) in the presence of 1 mM ABT or metyrapone. M-D was purified and identified as 3 β -hydroxy-11-oxo-olean-12-en-24,30-dioic acid by NMR and MS. Based on the structure of M-D, we propose the metabolic pathway of 18 β -GRA in strain G5.

Koldo Aurrekoetxea-Hernández, Elena Buetti. (Institute of Microbiology, University of Lausanne, CH-1011 Lausanne, Switzerland). Transforming Growth Factor β Enhances the Glucocorticoid Response of the Mouse Mammary Tumor Virus Promoter through Smad and GA-Binding Proteins. *Journal of Virology*, 78(5) (2004), 2201-2211.

Tissue-specific transcription is advantageously investigated by using viral promoters, which are selected for compact regulatory elements. Mouse mammary tumor virus (MMTV) has adapted to specialized cell types and targets initially B lymphocytes. We previously showed that, in B-cell lines, glucocorticoid-induced MMTV transcription requires an ETS family factor, GA-binding protein (GABP), bound in tandem to the MMTV DNA next to the glucocorticoid receptor (GR). We now report that transforming growth factor β (TGF- β) superinduces this response up to 10-fold through binding of its effectors, Smads, between the GABP-binding motifs. The basal level was unaffected. The TGF- β -glucocorticoid cooperation also depended

on GR and GABP binding, was transferable to another promoter, and occurred both with transiently transfected and with integrated templates. Smad3 associated in vitro with GR, with GABP α (via the MH2 domain), and with GABP β , Smad4 only with GABP α . Interactions of Smad3 with GABP (when coexpressed or endogenous to B cells) were shown by coprecipitation and by mammalian two-hybrid assay. This composite DNA element integrates three signaling pathways deriving from TGF- β , glucocorticoid hormones, and a unique ETS factor, and may allow MMTV to exploit factors from the milk. It may as well indicate novel possibilities for cellular regulatory networks.

M. Bavcon, P. Trebše, L. Zupančič-Kralj. (Laboratory for Environmental Research, Nova Gorica Polytechnic, Vipavska 13, P.O. Box 301, 5001-SI, Nova Gorica, Slovenia. Faculty of Chemistry and Chemical Technology, University of Ljubljana, Aškerčeva 5, 1000-SI, Ljubljana, Slovenia). Investigations of the determination and transformations of diazinon and malathion under environmental conditions using gas chromatography coupled with a flame ionisation detector. Chemosphere, 50(5) (2003), 595-601.

Degradation of two model insecticides, diazinon and malathion, and their degradation products 2-isopropyl-6-methyl-4-pyrimidinol--IMP (diazinon hydrolysis product) and malaoxon (malathion oxidation product) was compared and studied in the environment. The pesticides and their metabolites were extracted from samples (water, soil, chicory) with ethyl acetate and subsequently the extracts were analyzed by GC/FID. It was shown that hydrolysis is the major process in the degradation of these pesticides in water. In fact, 95% of diazinon was degraded, and only 10% of malathion was oxidised. In soil 30% of diazinon exposed to the sunlight was decomposed by photolysis, whereas in soil left in the darkness no degradation products were observed. In soil left under environmental conditions, 90% of diazinon was degraded and 40% from its initial concentration was transformed into IMP. The concentrations of the pesticides after 21 days on chicory were under maximal allowable concentration, which is 0.5 ppm for malathion and for diazinon. The concentration of malaoxon was more than twice as high as the allowable value, which is for the sum of malathion and malaoxon 3 ppm.

Manju Sharma, Rohini Sahni, Rekha Kansal, K R Koundal. Transformation of oilseed mustard Brassica juncea (L.) Czern & Coss cv. Pusajaisan with snowdrop lectin gene. Indian Journal of Biotechnology, 3 (2004), 97-102.

An efficient protocol has been developed to transfer snowdrop lectin gene (gna) to Brassica juncea (L.) Czern & Coss cv. Pusajaisan through Agrobacterium tumefaciens GV 2260 mediated transformation. High frequency regeneration of transformed plantlets has been achieved by using stem segments as explants. Analysis of the putative transformants by PCR amplification and Southern hybridization of genomic DNA showed the successful integration of the transgene in the nuclear genome. The transgenic plants will be further tested for bioassay in future.

María-Isabel Ramos-González, Arie Ben-Bassat, María-Jesús Campos, Juan L. Ramos. (Department of Biochemistry and Molecular and Cellular Biology of Plants, Estación Experimental del Zaidín, Consejo Superior de Investigaciones Científicas, 18008 Granada, Spain, Central Research and Development, DuPont Experimental Station, Wilmington, Delaware 19880-0328). Genetic Engineering of a Highly Solvent-Tolerant Pseudomonas putida Strain for Biotransformation of Toluene to p-Hydroxybenzoate. Applied and Environmental Microbiology, 69(9) (2003), 5216-5221.

The solvent-tolerant strain Pseudomonas putida DOT-T1E has been engineered for biotransformation of toluene into 4-hydroxybenzoate (4-HBA). P. putida DOT-T1E

transforms toluene into 3-methylcatechol in a reaction catalyzed by toluene dioxygenase. The *todC1C2* genes encode the α and β subunits of the multicomponent enzyme toluene dioxygenase, which catalyzes the first step in the Tod pathway of toluene catabolism. A DOT-T1EtodC mutant strain was constructed by homologous recombination and was shown to be unable to use toluene as a sole carbon source. The *P. putida* *pobA* gene, whose product is responsible for the hydroxylation of 4-HBA into 3,4-hydroxybenzoate, was cloned by complementation of a *Pseudomonas mendocina* *pobA1 pobA2* double mutant. This *pobA* gene was knocked out in vitro and used to generate a double mutant, DOT-T1EtodC*pobA*, that was unable to use either toluene or 4-HBA as a carbon source. The *tmo* and *pcu* genes from *P. mendocina* KR1, which catalyze the transformation of toluene into 4-HBA through a combination of the toluene 4-monoxygenase pathway and oxidation of *p*-cresol into the hydroxylated carboxylic acid, were subcloned in mini-Tn5Tc and stably recruited in the chromosome of DOT-T1EtodC*pobA*. Expression of the *tmo* and *pcu* genes took place in a DOT-T1E background due to cross-activation of the *tmo* promoter by the two-component signal transduction system TodST. Several independent isolates that accumulated 4-HBA in the supernatant from toluene were analyzed. Differences were observed in these clones in the time required for detection of 4-HBA and in the amount of this compound accumulated in the supernatant. The fastest and most noticeable accumulation of 4-HBA (12 mM) was found with a clone designated DOT-T1E-24.

Michael Seeger, Myriam González, Beatriz Cámara, Liliana Muñoz, Emilio Ponce, Lorenzo Mejías, Carolina Mascayano, Yesseny Vásquez, Silvia Sepúlveda-Boza. (Laboratorio de Microbiología Molecular y Biotecnología Ambiental, Departamento de Química, Universidad Técnica Federico Santa María, Valparaíso, Laboratorio de Química Médica y Ensayos Biológicos, Facultad de Ciencias Médicas, Universidad de Santiago de Chile, Santiago, Chile). Biotransformation of Natural and Synthetic Isoflavonoids by Two Recombinant Microbial Enzymes. Applied and Environmental Microbiology, 69(9) (2003), 5045-5050.

Isolation and synthesis of isoflavonoids has become a frequent endeavor, due to their interesting biological activities. The introduction of hydroxyl groups into isoflavonoids by the use of enzymes represents an attractive alternative to conventional chemical synthesis. In this study, the capabilities of biphenyl-2,3-dioxygenase (BphA) and biphenyl-2,3-dihydrodiol 2,3-dehydrogenase (BphB) of *Burkholderia* sp. strain LB400 to biotransform 14 isoflavonoids synthesized in the laboratory were investigated by using recombinant *Escherichia coli* strains containing plasmid vectors expressing the *bphA1A2A3A4* or *bphA1A2A3A4B* genes of strain LB400. The use of BphA and BphB allowed us to biotransform 7-hydroxy-8-methylisoflavone and 7-hydroxyisoflavone into 7,2',3'-trihydroxy-8-methylisoflavone and 7,3',4'-trihydroxyisoflavone, respectively. The compound 2'-fluoro-7-hydroxy-8-methylisoflavone was dihydroxylated by BphA at ortho-fluorinated and meta positions of ring B, with concomitant dehalogenation leading to 7,2',3'-trihydroxy-8-methylisoflavone. Daidzein (7,4'-dihydroxyisoflavone) was biotransformed by BphA, generating 7,2',4'-trihydroxyisoflavone after dehydration. Biotransformation products were analyzed by gas chromatography-mass spectrometry and nuclear magnetic resonance techniques.

Mokashi S.A., Paknikar K.M. Arsenic (III) oxidizing Microbacterium lacticum and its use in the treatment of arsenic contaminated groundwater. Letters in Applied Microbiology, 34(4) (2002), 258-262.

To develop a microbially-assisted process for the removal of arsenic from contaminated groundwater. A culture of *Microbacterium lacticum* oxidizing up to 50 mmol l⁻¹ arsenic (III) was isolated from municipal sewage by an enrichment culture technique. Using culture immobilized on brick pieces and packed in a glass column, complete oxidation of As (III)

from groundwater could be quickly achieved at neutral pH and ambient temperature with methanol as substrate. The oxidized As species were removed from groundwater using three different methods: zero valent iron, activated charcoal and ferric chloride. The oxidation of groundwater As (III) by a *M. lacticum*-immobilized column, followed by its removal using activated carbon, could be an efficient method for the treatment of As (III)-contaminated groundwater. The study will be useful in developing a combined microbiological-chemical process for treating arsenic-contaminated groundwater.

P. Pattanapitpaisal, N.L. Brown, L.E. Macaskie. (School of Biosciences, The University of Birmingham, Edgbaston B15 2TT, UK). Chromate reduction by *Microbacterium liquefaciens* immobilised in polyvinyl alcohol. *Biotechnology Letters*, 23(1) (2001), 61-65.

A polyvinyl alcohol-based immobilisation technique has been utilised for entrapping the newly-isolated chromate-reducing bacterium, *Microbacterium liquefaciens* MP30. Three immobilisation methods were evaluated: PVA-nitrate, PVA-borate and PVA-alginate. Chromate reduction was studied in batch and continuous-flow bioreactors, where the beads maintained integrity during continuous operation. PVA-borate and PVA-alginate cell beads showed a higher rate and extent of chromate reduction than PVA-nitrate cell beads in batch experiments. With the former 100 μM Cr(VI) was removed within 4 days, while only 40 μM Cr(VI) was removed using the latter, and with no increase in Cr(VI) removal subsequently. Cell activity was maintained during immobilisation but the rate of Cr(VI) removal by immobilised cells was only half that of an equivalent mass of free cells. Using PVA-alginate cell beads in a continuous-flow system, chromate removal was maintained at 90–95% from a 50 μM solution over 20 days without signs of bead breakdown.

Patil Y.B.; Paknikar K.M. Biodegradation of silver-cyanide from electroplating industry wastewater. *Letters in Applied Microbiology*, 30(1) (2000), 33-37.

A bacterial consortium capable of utilizing metal-cyanides as a source of nitrogen was used to develop a microbiological process for the detoxification of silver-cyanide from electroplating wastewater. When the treatment was carried out in a 27-l rotating biological contactor (RBC) in continuous mode, the system could achieve > 99.5% removal of 0.1 mmol l⁻¹ silver-cyanide (5 mg l⁻¹ cyanide and 10 mg l⁻¹ silver) in 10 h with sugarcane molasses (0.1 ml l⁻¹) as carbon source. The silver ions set free during biodegradation were efficiently adsorbed by the bacterial biomass. The RBC-treated effluent was found to be safe for discharge into the environment, as confirmed by chemical analysis and fish bioassay studies.

R. Schroll, F. Brahusi, U. Dörfler, S. Kühn, J. Fekete, J. C. Munch. (GSF--National Research Center for Environment and Health, Institute of Soil Ecology, D-85764, Neuherberg, Germany. Budapest University of Technology and Economics, Institute of General and Analytical Chemistry, H-111, Budapest, Hungary). Biomineralisation of 1,2,4-trichlorobenzene in soils by an adapted microbial population. *Environmental Pollution*, 127(3) (2004), 395-401.

In laboratory experiments the mineralisation of ¹⁴C-labelled 1,2,4-trichlorobenzene (1,2,4-TCB) in soils was studied by direct measurement of the evolved ¹⁴CO₂. The degradation capacity of the indigenous microbial population was investigated in an agricultural soil and in a soil from a contaminated site. Very low mineralisation of 1% within 23 days was measured in the agricultural soil. Whereas in the soil from the contaminated site the mineralisation occurred very fast and in high rates; up to 62% of the initially applied amount of 1,2,4-TCB were mineralised within 23 days. The transfer of the adapted microbial population into the agricultural soil significantly enhanced the mineralisation of 1,2,4-TCB in this soil, reflecting, that the transferred microbial population survived and maintained its

degradation ability in the new microbial ecosystem. Additional nutrition sources ((NH₄)₂HPO₄) increased the mineralisation rates in the first days significantly in the contaminated soil. In the soil from the contaminated site high amounts of non extractable ¹⁴C-residues were formed.

Authos Index

A. Bahrami, S.A. Shojaosadati, G. Moheballi.....	30
A. Binelli, A. Provini.....	12
A. C. Silva, M. Dezotti and G. L. Sant'Anna, Jr.	80
A. D'Annibale, R. Casa, F. Pieruccetti, M. Ricci, R. Marabottini.....	80
A. Jebanesan, M. R. Venkatachalam, G. Jagadeesan.	77
A. M. Omer, Y. Fadalla.....	66
A. P. Mitra, C. Sharma.....	95
A. P. Soldatkin, J. Montoriol, W. Santc, C. Martelet, N. Jaffrezic-Renaulta.....	91
A.K. Mukherjee, S.R.Mandal, UDITA Mandal AND B.C. Patra.	75
Aarti Mahajan, Suvasish Das.....	77
Abha Chhabra, S. Palria, V. K. Dadhwal.	66
Abraham F G Stevenson.....	73
Adolf Eisentraeger, Martin Schmidt, Hubertus Murrenhoff, Wolfgang Dott, Stefan Hahn.....	30
Agnes Pierwola, Tomasz Krupinski, Peter Zalupski, Michael Chiarelli, Domenic Castignetti.	30
Akio Tsuchii, Yutaka Tokiwa.....	31
Alfredo A. Marchetti, Mark G. Knize, Marina L. Chiarappa-Zucca, Ronald J. Pletcher and David W. Layton.	31
Alois Orlita.	32
Andrès Y, Texier A.C., Le Cloirec P.	12
Andrès Y.; Texier A.C.	32
Anil K.	91
Anthony Verdin , Anissa Lounès-Hadj Sahraoui, Roger Durand.....	32
Araceli Linares, Juan Manuel Caba, Francisco Ligero, Teresa de la Rubia, José artínez.....	81
Atul Kumar, Pallav Purohit, Santosh Rana and Tara Chandra Kandpal.	67

B. V. Chang, C. M. Yang, C. H. Cheng, S. Y. Yuan.	33
B.V. Chang, C.H. Yu, S.Y. Yuan.....	33
Bénédicte Viard, François Pihan, Sandrine Promeprat, Jean-Claude Pihan.....	12
Benoit Guieysse, Gunilla Viklund , Ann-Charlotte Toes, Bo Mattiasson.....	33
Benoit Van Aken, Jong Moon Yoon, Jerald L. Schnoor.....	34
Bernard Lachance, Agnès Y. Renoux, Manon Sarrazin , Jalal Hawari and Geoffrey I. Sunahara.	13
Bernd Marschnera, Karsten Kalbitzb.	34
Bethany M. McRae , Timothy M. LaPara and Raymond M. Hozalski.....	35
Bharat Bhushan, Louise Paquet,Jim C.Spain, Jalal Hawari.....	98
Bharat Bhushan, Sandra Trott, Jim C. Spain, Annamaria Halasz, Louise Paquet, Jalal Hawari.	98
Bhattacharya D, Mishra S, Sarma P M.....	73
Bill W. Bogan, Wendy R. Sullivan.	35
Bo Gejlsbjerg, Torben Madsen, Trine Thorup Andersen.	36
Brajesh K. Singh, Allan Walker, J. Alun W. Morgan, Denis J. Wright.	36
BridgwaterAV, Toft A J, and Brammer J G.	67
Buenaventurada P. Calabia, Yutaka Tokiwa.....	37
Byung-Taek Oh, Patrick J. Shea, Rhae A. Drijber, Galina K. Vasilyeva, Gautam Sarath.....	99
C. M. Kao, J. K. Liu, H. R. Lou, C. S. Lin, S. C. Chen.	99
C. Quintelas, T. Tavares.....	81
Carla A. Nicholson and Babu Z. Fathepure.....	37
Carlo Viti, Alessandra Pace, Luciana Giovannetti.....	81
Caroline Michela, Fabienne Battaglia-Bruneta, Canh Tran Minhb, Mireille Bruschi and Ioannis Ignatiadis.....	91
Céline Tixier, Martine Sancelme, Selim Aït-Aïssa, Pascale Widehem, Frédérique Bonnemoy, Annie Cuer, Nicole Truffaut, Henri Veschambre.	100

César Ricardo Teixeira Tarley, Marco Aurélio Zezzi Arruda.....	13
Chaisuksant Y.....	13
Chang-Tang Chang, Bor-Yann Chen, I-Shing Shiu, Fu-Teng Jeng.	14
Christoph Bausch, Matthew Ramsey, Tyrrell Conway.	67
Chunlong Zhang, Joseph B. Hughes.....	37
Costantino Vischetti, Ettore Capri, Marco Trevisan, Cristiano Casucci, Piero Perucci.	14
Cynthia Davis, Todd Cort, Dongping Dai, Tissa H. Illangasekare, Junko Munakata-Marr.....	38
D. G. Heijerick, C. R. Janssen, C. Karlèn, I. Odnevall Wallinder, C. Leygraf.....	14
D. Moldes, S. Rodríguez Couto, C. Cameselle, M. A. Sanromán.	38
D. N. Mishra, K. Kumar.....	78
D.Y. Bojinova, R.G. Velkova.....	100
Dai Kitamoto, Toru Ikegami, Gaby Tiemi Suzuki, Akira Sasaki, Yu-ichiro Takeyama, Yasushi Idemoto, Nobuyuki Koura, Hiroshi Yanagishita.....	101
Daljít S. Arora, Paramjít K. Gill.....	74
.Daljít Singh Arora, Mukesh Chander and Paramjít Kaur Gill.	39
Daljít Singh Arora, Paramjít Kaur Gill.	74
Daniel Filippini, Tony P. M. Anderssonb, Samuel P. S. Svenssonb, Ingemar Lundströma....	92
Darryl P. Arfsten, Dennis T. Burton, Daniel J. Fisher, John Callahan, Cody L. Wilson, Kenneth R. Still, Barry J. Spargo.	39
David J. Hopper, Lisa Cottrell.....	101
Diane Fournier, Annamaria Halasz, Jim Spain, Ronald J. Spangord, Jeffrey C. Bottaro, Jalal Hawari.....	39
Dogru M, Howarth C R, Akay G, Keskinler B, Malik A A.....	68
Dong W. Kim, Daniel K. Cha, J. Wang, C. P. Huang..	82
E. G. Gregoricha, M. H. Beareb, U. Stoklasa, P. St-Georges.	40
Elizalde-González M.P., Peláez-Cid A.A.....	41
Epaminondas Voutsas, Kostis Magoulas, Dimitrios Tassios.....	15

Fabio Fava, Silvia Gentilucci, Giulio Zanaroli.	41
Farinazleen Mohamad Ghazali , Raja Noor Zaliha Abdul Rahman, Abu Bakar Salleh and Mahiran Basri.	42
Francisco Jurado, Antonio Canoa, José Carpiob.	68
G. Chen, J. Andries, H. Spliethoff.	68
Geremias R., Pedrosa R.C., Benassi J.C., Fávere V.T., Stolberg J.,Menezes C.T.B., Laranjeira M.C.M.	82
Gerhardt F. Riedel, Nathalie Valette-Silver.....	15
God B. T	69
Graham H. Crossa, Andrew A. Reevesa, Stuart Branda, Jonathan F. Popplewellb, Louise L. Peelb, Marcus J. Swannb, Neville J. Freeman.	92
H. Kang, S.Y. Hwang, Y.M. Kim, E. Kim, Y.-S. Kim, S.-K. Kim, S.W. Kim, C.E. Cerniglia, K.L. Shuttleworth, and G.J. Zylstra.	42
H. Rezai, F. M. Yusoff, A. Kawamura, A. Arshad, B. H. R. Othman.	69
Hai Yan, Gang Pan.....	16
Hai Yan, Gang Pan.....	16
Hangzhou.....	16
Henry H. Tabak, Richard Scharp, John Burckle, Fred K. Kawahara, Rakesh Govind..	101
Hillol Guha, Krishnaswamy Jayachandran, Florentin Maurrassé.	82
I.J. Díaz-Ramírez, H. Ramírez-Saad, M. Gutiérrez-Rojas, E. Favela-Torres.	42
Inge S. Fomsgaard, Anne G. Mortensen, Sandra C. K. Carlsen..	102
Ingle ST, Pachpande B G, Mali D S.	96
.Isabel Sierra, José Luis Valera, M. Luisa Marina, and Fernando Laborda.	43
Iveta Řezníčková, Jaromír Hoffmann, Karel Komárek.	43
J. A. Fernández, A. Carballeira.	17
J. Taube, K. Vorkamp, M. Förster, R. Herrmann.	78
J. W. C. Wong, L. Xiang, X. Y. Gu, L. X. Zhou.	17

J. W. Goodrum.....	69
Jadwiga Szostak-Kotowa.....	43
Jae Jun Jeong, Ji Hyun Kim, Chi-Kyung Kim, Ingyu Hwang and Kyoung Lee.....	44
Jaka Widada, Hideaki Nojiri, Takako Yoshida, Hiroshi Habe, Toshio Omori.	44
Jan Erik Mattsson, Pieter D. Kofman.	69
Jens Dittmann, Wolfgang Heyser and Heike Bücking.....	45
Jeong-Hun Park, Yucheng Feng, Pingsheng Ji, Thomas C. Voice, Stephen A. Boyd.....	18
Jiantao Pang, Chunhai Fan, Xinjian Liu, Ting Chen, Genxi Li.....	93
Ji-Guang Gu, Yanzhen Fan, Ji-Dong Gu.	45
Joanita Coelho, C. U. Rivonkar, N. S. Bhavesh., M. Jothi & U. M. X. Sangodkar..	46
Jörg Overhage, Alexander Steinbüchel, Horst Priefert.....	102
Jukka Ahtiainen, Miia Aalto, Piia Pessala.....	46
Julio A. (Departamento Interuniversitario de Ecología, Edificio de Ciencias, Universidad de Alcalá, Alcalá de Henares, Madrid E-28871, Spain). Fluoride toxicity to aquatic organisms: a review. <i>Chemosphere</i> , 50(3), (2003), 251-264.	18
Jung-Hyun Lee, Seong-Young Jung, Sang-Jin Kim.	47
K Murugesan.	83
K. Kalbitz, J. Schmerwitz, D. Schwesig, E. Matzner.....	47
K. L. Londry, L. L. Jahnke, D. J. Des Marais..	75
K. Padma Dorothy, B. Satyanarayana, C. Kalavati, A. V. Raman, F. Dehairs.	48
Kaijun Wang and Baoshan Xing.....	19
Kamlesh Kanwar, Anju Bhardwaj, Sandhya Agarwal & D R Sharma.	103
Kari T. Steffen, Annele Hatakka, Martin Hofrichter.....	48
Katrine Borgå, Bjørn Gulliksen, Geir Wing Gabrielsen, Janneche Utne Skaare.....	19
Kaushik N.....	78
Kazunari Sei ¹ , Yoshiro Sugimoto ¹ , Kazuhiro Mori ¹ , Hideaki Maki, Tetsuro Kohno.	49
Keishiro Yoshida, Kazuo Furihata, Hiroshi Habe, Hisakazu Yamane, Toshio Omori.....	103

Kelly P. Nevin, Kevin T. Finneran, Derek R. Lovley.	83
Koldo Aurrekoetxea-Hernández, Elena Buetti.	103
Krassimira Hristova, Binyam Gebreyesus, Douglas Mackay, and Kate M. Scow.	49
Kumiko Miyoshi, Takehiko Nishio, Akio Yasuhara, Masatoshi Morita, Takayuki Shibamoto. .	20
Kwang-Soup Song, Toshikatsu Sakaia, b, Hirofumi Kanazawaa, Yuta Arakia, Hitoshi Umezawaa, Minoru Tachikia, Hiroshi Kawaradaa.	93
L.J.R. Foster, A. Saufi, P.J. Holden.	75
Lal B, Mishra S, Bhattacharya D, Sarma P M.	96
Lal B, Mishra S, Ramesh KC Jyot J.	83
Lance D. Hansen, Cathy Nestler, Dave Ringelberg, Rakesh Bajpai. .	84
Lars Elsgaard, Giulio Pojana, Tommaso Miraval, Jørgen Eriksen, Antonio Marcomini.	49
Le Floch S., Guyomarch J., Merlin F., Børseth J.F., Le Corre P., Lee K.	85
Leonóra Száraz, Judit Becznerb.	50
Lepo J.E., Cripe C.R., Kavanaugh J.L., Zhang S., Norton G.P.	50
Lourenço N.D., Novais J.M., Pinheiro H.M.	51
Luciana Migliore, Salvatore, Cozzolino, Maurizio Fiori.	20
Luis A. Rios-Hernandez, Lisa M. Gieg, Joseph M. Suflita.	51
M. Bavcon, P. Trebše, L. Zupančič-Kralj.	104
M. Cerón-Rivera, M. M. Dávila-Jiménez, M. P. Elizalde-González.	52
M. Fürhacker, A. Pressl, R. Allabashi.	52
M. Humar, M. Bokan, S. A. Amartey, M. entjurc, P. Kalan, F. Pohleven.	85
.M. Schwenk, T. Gabrio, O. Pöpke, T. Wallenhorst.	93
M. Trigui, S. Pulvin, P. Poupin, and D. Thomas.	52
M.R. Kosseva, C.A. Kent, D.R. Lloyd.	85
Mahnaz Mazaheri Assadi, Maryam Mazaheri, Taher Nejad Satari, Ashrafulsadat Noohi, Manoochehr Shahamat, Morris Levin.	20
Manju Sharma, Rohini Sahni, Rekha Kansal, K R Koundal.	104

María-Isabel Ramos-González, Arie Ben-Bassat, María-Jesús Campos, Juan L. Ramos.....	104
Mark J. Hopkins, Hans N. Englyst, Sandra Macfarlane, Elizabeth Furrie, George T. Macfarlane, Andrew J. McBain.....	53
Maurizio Aceto, Ornella Abollino, Raffaele Conca, Mery Malandrino, Edoardo Mentasti, Corrado Sarzanini.....	76
Mayra A. Laraa, Antonio J. Rodríguez-Malaverb, Orlando J. Rojas, Otón Holmquistc, Aura M. González, Johnny Bullóna, Nancy Peñalozab, Elisa Araujoa.	53
Meltem Urgun-Demirtas, Krishna R. Pagilla, Benjamin C. Stark, Dale Webster.	54
Merja Itävaara, Sari Karjomaa, Johan-Fredrik Selin.	54
Meshram J R.....	70
Michael F. Coughlin, Brian K. Kinkle, Paul L. Bishop.	54
Michael Seeger, Myriam González, Beatriz Cámara, Liliana Muñoz, Emilio Ponce, Lorenzo Mejías, Carolina Mascayano, Yesseny Vásquez, Silvia Sepúlveda-Boza.	105
Mikael Eriksson, Erik Sodersten, Zhongtang Yu, Gunnel Dalhammar, William W. Mohn.....	55
Mishra S, Jyot J, Kuhad R C, Lal B	86
Mishra S, Jyot J.....	56
Mohindru V K, Singh Kawaljeet, Jena B K, Siddiqui N A, Kulkarni A G.....	96
Mokashi S.A.,Paknikar K.M.	105
Mónica João de Barros Amorim, José Paulo Sousa, António J. A. Nogueira, Amadeu M. V. M. Soares.	21
N. Perron, U. Welander.	56
Nakamura N., Nakano K., Sugiura N., Matsumura M.....	56
Naoto Ogawa, Hideo Okamura, Hirofumi Hirai, Tomoaki Nishida.....	57
Nico Boon, Eva M. Top, Willy Verstraete, Steven D. Siciliano.....	21
Nirmala Agrawal, R.S. Sachan.....	86
Nirupama Mallick.....	86
Norbert Scholl.....	57
Ojumu, T.V., Yu, J. and Solomon, B.O.	57

Olivier Potin , Catherine Rafin, Etienne Veignie.	87
Omry Koren, Vishnia Knezevic, Eliora Z. Ron, Eugene Rosenberg	87
P. Adamo, M. Arienzo, M. Pugliese, V. Roca, P. Violante.	22
P. C. J. Roach, D. K. Ramsdena, J. Hughesb, P. Williamsa.	94
P. Gill, D. Arora.....	74
P. Guiraud, D. Villemain, M. Kadri, O. Bordjiba, R. Steiman.	58
P. Pattanapitpaisal, N.L. Brown, L.E. Macaskie.....	106
P. Sudhaa, H. I. Somashekharb, Sandhya Raoc, N. H. Ravindranath.....	70
Pairat Kaewsarn.....	22
Patil Y.B.; Paknikar K.M..	106
Patricia A. Terry, Wendy Stone	23
Peter F. Landrum, Guilherme R. Lotufo, Duane C. Gossiaux, Michelle L. Gedeon, Jong-Hyeon Lee.....	23
PK Gill, DS Arora, M Chander.....	24
Prem Kishore, A. K. Dikshit and Rajendra Kumar.....	79
R. Bennicelli, Z. Stępniewska, A. Banach, K. Szajnocha, J. Ostrowski.	87
R. Borja, B. Rincón, F. Raposo, E. Sánchez, A. Martín.	58
R. Margesin, D. Labbé, F. Schinner, C. W. Greer, L. G. Whyte.....	59
R. Nandakumar, S. Babu, R. Viswanathan, J. Sheela, T. Raguchander , R. Samiyappan. ...	79
R. Schroll, F. Brahushi, U. Dörfler, S. Kühn, J. Fekete, J. C. Munch.....	106
R. Sunil Kumar.	70
Rajeshwari V, Kusum Lata, Pant D C, Kishore V V N.	71
René van Herwijnen, Dirk Springael, Pieter Slot, Harrie A. J. Govers, John R. Parsons. (Richard Wasserbauer.....	59
Riina Turpeinen, Timo Kairesalo, Max M. Häggblom.	88
Rincón N.; Chacín E., Marín J., Torrijos M., Moletta R., Fernández N.	60
S. Dapía, V. Santos and J. C. Parajó.	71

S. Hwang and T. J. Cutright.....	60
S. Janarthanan, P. Suresh.	79
S. K. Singh, A. K. Dikshit, K. D. Srivastava, R. S. Tanwar, P. Dureja.....	80
S. Lecoeur, B. Videmann, Ph. Berny.	76
S. Nonhebel.....	71
Sandra Domenek, Pierre Feuilleley, Jean Gratraud, Marie-Hélène Morel, Stéphane Guilbert.	60
Sandra Trott, Shirley F. Nishino, Jalal Hawari, Jim C. Spain.	61
Seiji Katano, Yuka Matsuo, Ken'ichi Hanaoka.	24
Seiji Shibasakia, Atsuo Tanakab, Mitsuyoshi Ueda..	94
Shardendu, N. Salhani, S. F. Boulyga, E. Stengel.	88
Shen-Yi Chen, Jih-Gaw Lin	89
Sinha S and Sreekesh S.....	97
Sinha S, Saxena R, and Singh S.....	24
Sirpa Metsärinne, Päivi Rantanen, Reijo Aksela, Tuula Tuhkanen.....	61
Smith K R. Uma R, Zhang J, Kishore V, V N, Joshi V.	72
Solange In [^] es Mussatto, In [^] es Conceição Roberto.	89
Srinivas S N.	72
Sugijanto Kartosentono, Ana Nuraida, Gunawan Indrayanto, Noor Cholies Zaini.....	89
Suhendrayatna, Akira Ohki, Tsunenori Nakajima, Shigeru Maeda.	25
Surinder Katyal, Kelly Thambimuthu, Marjorie Valix.....	72
Sven Burreau, Yngve Zebühr, Dag Broman, Rasha Ishaq.	25
T. Manios, E. I. Stentiford, P. Millner.....	26
T. Srinath, T. Verma, P. W. Ramteke, S. K. Garg.....	26
Tatsuya Ueki, Yasuhisa Sakamoto, Nobuo Yamaguchi, and Hitoshi Michibata.	27
Tetsuo Ando, Megumi Yamamoto, Takashi Tomiyasu, Jun Hashimoto, Tomoyuki Miura, Atsuhiko Nakano, Suminori Akiba.	27

Thomas Borch, Per Ambus, Frank Laturus, Bo Svensmark, Christian Grøn.	62
Thomas Pümpel, Lynne E. Macaskie, John A. Finlay, Ludo Diels, Marios Tsezos	89
Tiina Petänen, Martin Romantschuk.	28
Tomonori Ishigaki, Wataru Sugano, Akane Nakanishi, Masafumi Tateda, Michihiko Ike, Masanori Fujita.	62
U. F. Wehmeier, W. Piepersberg.....	97
V. Matus, M. A. Sánchez, M. Martínez, B. González.....	63
Volker Riis, Sabine Kleinsteuber, and Wolfgang Babel.	63
Volker Riis, Wolfgang Babel, Oscar Héctor Pucci.	64
W. J. Reynolds, S. W. Feist, G. J. Jones, B. P. Lyons, D. A. Sheahan, G. D. Stentiford.....	76
W. M. Law, W. N. Lau, K. L. Lo, L. M. Wai, S. W. Chiu.....	90
Winnie Dejonghe, Ellen Berteloot, ¹ Johan Goris, Nico Boon, Katrien Crul, Siska Maertens, Monica Höfte, Paul De Vos, Willy Verstraete, Eva M. Top.	64
Xiangchun Quan, Hanchang Shi, Jianlong Wang, Yi Qian.	64
Xiaoqi Zhang, Paul L. Bishop.....	65
Xin Hu, Zhuhong Ding, Yijun Chen, Xiaorong Wang, Lemei Dai.	28
Y. X. Chen, Q. Lin, Y. M. Luo, Y. F. He, S. J. Zhen, Y. L. Yu, G. M. Tian, M. H. Wong.	90
Yang Liu, Xiao Yu, Rui Zhao, Di-Hua Shangguan, Zuyi Bo, Guoquan Liu.	94
Young Soo Keum and Qing X. Li.	65
Youn-Seok Kang, Masumi Yamamuro, Shigeki Masunaga, Junko Nakanishi.....	28
Yuh-Ming Uanga, Tse-Chuan Chou.....	95
Z. H. Ye, A. J. M. Baker, M. H. Wong, A. J. Willis.	29
Zhanpeng Jiang, Hongwei Yang, Lixin Sun, Shaoqi Shi.....	66
Zümriye Aksu, Gönül Dönmez.....	29

Name of Journals

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A. Binelli, A. Provini.....	28
Andrès Y, Texier A.C., Le Cloirec P.	28
Bénédicte Viard, François Pihan, Sandrine Promeprat, Jean-Claude Pihan.....	28
Bernard Lachance, Agnès Y. Renoux, Manon Sarrazin , Jalal Hawari and Geoffrey I. Sunahara.	29
César Ricardo Teixeira Tarley, Marco Aurélio Zezzi Arruda.....	29
Chaisuksant Y.....	29
Chang-Tang Chang, Bor-Yann Chen, I-Shing Shiu, Fu-Teng Jeng.	30
Costantino Vischetti, Ettore Capri, Marco Trevisan, Cristiano Casucci, Piero Perucci.	30
D. G. Heijerick, C. R. Janssen, C. Karlèn, I. Odnevall Wallinder, C. Leygraf.....	31
Epaminondas Voutsas, Kostis Magoulas, Dimitrios Tassios.....	31
Gerhardt F. Riedel, Nathalie Valette-Silver.....	31
Hai Yan, Gang Pan.....	32
Hai Yan, Gang Pan.....	32
Hangzhou.....	33
J. A. Fernández, A. Carballeira.	33
J. W. C. Wong, L. Xiang, X. Y. Gu, L. X. Zhou.	33
Jeong-Hun Park, Yucheng Feng, Pingsheng Ji, Thomas C. Voice, Stephen A. Boyd.....	34
Julio A. (Departamento Interuniversitario de Ecología, Edificio de Ciencias, Universidad de Alcalá, Alcalá de Henares, Madrid E-28871, Spain). Fluoride toxicity to aquatic organisms: a review. <i>Chemosphere</i> , 50(3), (2003), 251-264.	35
Kaijun Wang and Baoshan Xing.....	35
Katrine Borgå, Bjørn Gulliksen, Geir Wing Gabrielsen, Janneche Utne Skaare.....	35
Luciana Migliore, Salvatore, Cozzolino, Maurizio Fiori.	36
Mahnaz Mazaheri Assadi, Maryam Mazaheri, Taher Nejad Satari, Ashrafulsadat Noohi, Manoochehr Shahamat, Morris Levin.	36
Kumiko Miyoshi, Takehiko Nishio, Akio Yasuhara, Masatoshi Morita, Takayuki Shibamoto..	36
Mónica João de Barros Amorim, José Paulo Sousa, António J. A. Nogueira, Amadeu M. V. M. Soares. (Departamento de Biologia, Universidade de Aveiro, P-3810-193, Aveiro, Portugal. Instituto do Ambiente e Vida--Dpto. Zoologia da Universidade de Coimbra, P-3004-517, Coimbra, Portugal). Bioaccumulation and elimination of ¹⁴ C-lindane by <i>Enchytraeus albidus</i> in artificial (OECD) and a natural soil. <i>Chemosphere</i> , 49(3) (2002), 323-329.....	37
Nico Boon, Eva M. Top, Willy Verstraete, Steven D. Siciliano.....	37
P. Adamo, M. Arienzo, M. Pugliese, V. Roca, P. Violante.	38
Pairat Kaewsarn.....	39
Patricia A. Terry, Wendy Stone	39

Peter F. Landrum, Guilherme R. Lotufo, Duane C. Gossiaux, Michelle L. Gedeon, Jong-Hyeon Lee.....	39
PK Gill, DS Arora, M Chander.....	40
Seiji Katano, Yuka Matsuo, Ken'ichi Hanaoka.	40
Sinha S, Saxena R, and Singh S.....	41
Suhendrayatna, Akira Ohki, Tsunenori Nakajima, Shigeru Maeda.	41
Sven Burreau, Yngve Zebühr, Dag Broman, Rasha Ishaq.	41
T. Manios, E. I. Stentiford, P. Millner.....	42
T. Srinath, T. Verma, P. W. Ramteke, S. K. Garg.....	42
Tatsuya Ueki, Yasuhisa Sakamoto, Nobuo Yamaguchi, and Hitoshi Michibata.	43
Tetsuo Ando, Megumi Yamamoto, Takashi Tomiyasu, Jun Hashimoto, Tomoyuki Miura, Atsuhiko Nakano, Suminori Akiba.	43
Tiina Petänen, Martin Romantschuk.	44
Xin Hu, Zhuhong Ding, Yijun Chen, Xiaorong Wang, Lemei Dai.	44
Youn-Seok Kang, Masumi Yamamuro, Shigeki Masunaga, Junko Nakanishi.....	45
Z. H. Ye, A. J. M. Baker, M. H. Wong, A. J. Willis.	45
Zümriye Aksu, Gönül Dönmez.....	46
A. Bahrami, S.A. Shojaosadati, G. Moheballi.....	46
Adolf Eisentraeger, Martin Schmidt, Hubertus Murrenhoff, Wolfgang Dott, Stefan Hahn.....	46
Agnes Pierwola, Tomasz Krupinski, Peter Zalupski, Michael Chiarelli, Domenic Castignetti.	47
Akio Tsuchii, Yutaka Tokiwa.....	47
Alfredo A. Marchetti, Mark G. Knize, Marina L. Chiarappa-Zucca, Ronald J. Pletcher and David W. Layton.	47
Alois Orlita.	48
Andrès Y.; Texier A.C.	48
Anthony Verdin , Anissa Lounès-Hadj Sahraoui, Roger Durand.....	48
B.V. Chang, C.H. Yu, S.Y. Yuan.....	49
B. V. Chang, C. M. Yang, C. H. Cheng, S. Y. Yuan.	49
Benoit Guieysse, Gunilla Viklund , Ann-Charlotte Toes, Bo Mattiasson.....	49
Benoit Van Aken, Jong Moon Yoon, Jerald L. Schnoor.....	50
Bernd Marschnera, Karsten Kalbitzb.	50
Bethany M. McRae , Timothy M. LaPara and Raymond M. Hozalski.....	51
Bill W. Bogan, Wendy R. Sullivan.	51
Bo Gejlsbjerg, Torben Madsen, Trine Thorup Andersen.	52
Brajesh K. Singh, Allan Walker, J. Alun W. Morgan, Denis J. Wright.	52

Buenaventurada P. Calabia, Yutaka Tokiwa.....	53
Carla A. Nicholson and Babu Z. Fathepure.....	53
Chunlong Zhang, Joseph B. Hughes.....	53
Cynthia Davis, Todd Cort, Dongping Dai, Tissa H. Illangasekare, Junko Munakata-Marr.....	54
D. Moldes, S. Rodríguez Couto, C. Cameselle, M. A. Sanromán.	54
Daljit Singh Arora, Mukesh Chander and Paramjit Kaur Gill.	55
Darryl P. Arfsten, Dennis T. Burton, Daniel J. Fisher, John Callahan, Cody L. Wilson, Kenneth R. Still, Barry J. Spargo.	55
Diane Fournier, Annamaria Halasz, Jim Spain, Ronald J. Spangord, Jeffrey C. Bottaro, Jalal Hawari.....	56
E. G. Gregoricha, M. H. Beareb, U. Stoklasa, P. St-Georges.	56
Elizalde-González M.P., Peláez-Cid A.A.....	57
Fabio Fava, Silvia Gentilucci, Giulio Zanaroli.	57
Farinazleen Mohamad Ghazali , Raja Noor Zaliha Abdul Rahman, Abu Bakar Salleh and Mahiran Basri.	58
H. Kang, S.Y. Hwang, Y.M. Kim, E. Kim, Y.-S. Kim, S.-K. Kim, S.W. Kim, C.E. Cerniglia, K.L. Shuttleworth, and G.J. Zylstra.	58
I.J. Díaz-Ramírez, H. Ramírez-Saad, M. Gutiérrez-Rojas, E. Favela-Torres.	58
Isabel Sierra, José Luis Valera, M. Luisa Marina, and Fernando Laborda.	59
Iveta Řezníčková, Jaromír Hoffmann, Karel Komárek.	59
Jadwiga Szostak-Kotowa.....	60
Jae Jun Jeong, Ji Hyun Kim, Chi-Kyung Kim, Ingyu Hwang and Kyoung Lee.....	60
Jaka Widada, Hideaki Nojiri, Takako Yoshida, Hiroshi Habe, Toshio Omori.	60
Jens Dittmann, Wolfgang Heyser and Heike Bücking.....	61
Ji-Guang Gu, Yanzhen Fan, Ji-Dong Gu.	61
Joanita Coelho, C. U. Rivonkar, N. S. Bhavesh., M. Jothi & U. M. X. Sangodkar..	62
Jukka Ahtiainen, Miia Aalto, Piia Pessala.....	62
Jung-Hyun Lee, Seong-Young Jung, Sang-Jin Kim.	63
K. Kalbitz, J. Schmerwitz, D. Schwesig, E. Matzner.....	63
K. Padma Dorothy, B. Satyanarayana, C. Kalavati, A. V. Raman, F. Dehairs.	64
Kari T. Steffen, Annele Hatakka, Martin Hofrichter.....	64
Kazunari Sei1, Yoshiro Sugimoto1, Kazuhiro Mori1, Hideaki Maki, Tetsuro Kohno.	65
Krassimira Hristova, Binyam Gebreyesus, Douglas Mackay, and Kate M. Scow.....	65
Lars Elsgaard, Giulio Pojana, Tommaso Miraval, Jørgen Eriksen, Antonio Marcomini.....	66
Leonóra Száraz, Judit Becznerb.....	66
Lepo J.E., Cripe C.R., Kavanaugh J.L., Zhang S., Norton G.P.	66

Lourenço N.D., Novais J.M., Pinheiro H.M.	67
Luis A. Rios-Hernandez, Lisa M. Gieg, Joseph M. Suflita.	67
M. Cerón-Rivera, M. M. Dávila-Jiménez, M. P. Elizalde-González.....	68
M. Fürhacker, A. Pressl, R. Allabashi.....	68
M. Trigui, S. Pulvin, P. Poupin, and D. Thomas.	69
Mark J. Hopkins, Hans N. Englyst, Sandra Macfarlane, Elizabeth Furrie, George T. Macfarlane, Andrew J. McBain.....	69
Mayra A. Laraa, Antonio J. Rodríguez-Malaverb, Orlando J. Rojas, Otón Holmquistc, Aura M. Gonzáleza, Johnny Bullóna, Nancy Peñalozab, Elisa Araujoa.	69
Meltem Urgun-Demirtas, Krishna R. Pagilla, Benjamin C. Stark, Dale Webster.	70
Merja Itävaara, Sari Karjomaa, Johan-Fredrik Selin.	70
Michael F. Coughlin, Brian K. Kinkle, Paul L. Bishop.	71
Mikael Eriksson, Erik Sodersten, Zhongtang Yu, Gunnel Dalhammar, William W. Mohn.....	71
Mishra S, Jyot J.....	72
N. Perron, U. Welander.	72
Nakamura N., Nakano K., Sugiura N., Matsumura M.....	73
Naoto Ogawa, Hideo Okamura, Hirofumi Hirai, Tomoaki Nishida.....	73
Norbert Scholl.....	73
Ojumu, T.V., Yu, J. and Solomon, B.O.	74
P. Guiraud, D. Villemain, M. Kadri, O. Bordjiba, R. Steiman.	74
R. Borja, B. Rincón, F. Raposo, E. Sánchez, A. Martín.	74
R. Margesin, D. Labbé, F. Schinner, C. W. Greer, L. G. Whyte.....	75
René van Herwijnen, Dirk Springael, Pieter Slot, Harrie A. J. Govers, John R. Parsons. (Richard Wasserbauer.....	76
Rincón N.; Chacín E., Marín J., Torrijos M., Moletta R., Fernández N.	76
S. Hwang and T. J. Cutright.....	76
Sandra Domenek, Pierre Feuilloley, Jean Gratraud, Marie-Hélène Morel, Stéphane Guilbert.	77
Sandra Trott, Shirley F. Nishino, Jalal Hawari, Jim C. Spain.	77
Sirpa Metsärinne, Päivi Rantanen, Reijo Aksela, Tuula Tuhkanen.....	77
Thomas Borch, Per Ambus, Frank Laturus, Bo Svensmark, Christian Grøn.	78
Tomonori Ishigaki, Wataru Sugano, Akane Nakanishi, Masafumi Tateda, Michihiko Ike, Masanori Fujita.	78
V. Matus, M. A. Sánchez, M. Martínez, B. González.....	79
Volker Riis, Sabine Kleinsteuber, and Wolfgang Babel.	79
Volker Riis, Wolfgang Babel, Oscar Héctor Pucci.....	80

Winnie Dejonghe, Ellen Berteloot, ¹ Johan Goris, Nico Boon, Katrien Crul, Siska Maertens, Monica Höfte, Paul De Vos, Willy Verstraete, Eva M. Top.	80
Xiangchun Quan, Hanchang Shi, Jianlong Wang, Yi Qian.	81
Xiaoqi Zhang, Paul L. Bishop.....	81
Young Soo Keum and Qing X. Li.....	81
Zhanpeng Jiang, Hongwei Yang, Lixin Sun, Shaoqi Shi.....	82
A. M. Omer, Y. Fadalla.	82
Abha Chhabra, S. Palria, V. K. Dadhwal.	82
Atul Kumar, Pallav Purohit, Santosh Rana and Tara Chandra Kandpal.	83
BridgwaterAV, Toft A J, and Brammer J G.	83
Christoph Bausch, Matthew Ramsey, Tyrrell Conway.	83
Dogru M, Howarth C R, Akay G, Keskinler B, Malik A A.....	84
Francisco Jurado, Antonio Canoa, José Carpiob.	84
G. Chen, J. Andries, H. Spliethoff.	85
God B. T	85
H. Rezai, F. M. Yusoff, A. Kawamura, A. Arshad, B. H. R. Othman.	85
J. W. Goodrum.....	85
Jan Erik Mattsson, Pieter D. Kofman.	86
Meshram J R.....	86
P. Sudhaa, H. I. Somashekharb, Sandhya Raoc, N. H. Ravindranath.....	86
R. Sunil Kumar.	87
Rajeshwari V, Kusum Lata, Pant D C, Kishore V V N.	87
S. Dapía, V. Santos and J. C. Parajó.	87
S. Nonhebel.....	88
Smith K R. Uma R, Zhang J, Kishore V, V N, Joshi V.	88
Srinivas S N.	88
Surinder Katyal, Kelly Thambimuthu, Marjorie Valix.....	89
Abraham F G Stevenson.....	89
Bhattacharya D, Mishra S, Sarma P M.....	90
Daljit S. Arora, Paramjit K. Gill.....	90
Daljit Singh Arora, Paramjit Kaur Gill.	90
P. Gill, D. Arora.....	90
A.K. Mukherjee, S.R.Mandal, UDITA Mandal AND B.C. Patra.	91
K. L. Londry, L. L. Jahnke, D. J. Des Marais..	91
L.J.R. Foster, A. Saufi, P.J. Holden.....	92

Maurizio Aceto, Ornella Abollino, Raffaele Conca, Mery Malandrino, Edoardo Mentasti, Corrado Sarzanini.....	92
S. Lecoeur, B. Videmann, Ph. Berny.	92
W. J. Reynolds, S. W. Feist, G. J. Jones, B. P. Lyons, D. A. Sheahan, G. D. Stentiford.....	93
A. Jebanesan, M. R. Venkatachalam, G. Jagadeesan.	93
Aarti Mahajan, Suvasish Das.....	93
D. N. Mishra, K. Kumar..	94
J. Taube, K. Vorkamp, M. Förster, R. Herrmann.	94
Kaushik N.....	94
Prem Kishore, A. K. Dikshit and Rajendra Kumar.....	95
R. Nandakumar, S. Babu, R. Viswanathan, J. Sheela, T. Raguchander , R. Samiyappan.	95
S. Janarthanan, P. Suresh.	96
S. K. Singh, A. K. Dikshit, K. D. Srivastava, R. S. Tanwar, P. Dureja.....	96
A. C. Silva, M. Dezotti and G. L. Sant'Anna, Jr.	96
A. D'Annibale, R. Casa, F. Pieruccetti, M. Ricci, R. Marabottini.....	97
Araceli Linares, Juan Manuel Caba, Francisco Ligeró, Teresa de la Rubia, José artínez.....	97
C. Quintelas, T. Tavares.....	97
Carlo Viti, Alessandra Pace, Luciana Giovannetti.....	97
Dong W. Kim, Daniel K. Cha, J. Wang, C. P. Huang..	98
Geremias R., Pedrosa R.C., Benassi J.C., Fávère V.T., Stolberg J., Menezes C.T.B., Laranjeira M.C.M.	98
Hillol Guha, Krishnaswamy Jayachandran, Florentin Maurasse.	99
K Murugesan.	99
Kelly P. Nevin, Kevin T. Finneran, Derek R. Lovley.	99
Lal B, Mishra S, Ramesh KC Jyot J.....	99
Lance D. Hansen, Cathy Nestler, Dave Ringelberg, Rakesh Bajpai..	100
Le Floch S., Guyomarch J., Merlin F., Børseth J.F., Le Corre P., Lee K.	101
M. Humar, M. Bokan, S. A. Amartey, M. entjurc, P. Kalan, F. Pohleven.	101
M.R. Kosseva, C.A. Kent, D.R. Lloyd.	102
Mishra S, Jyot J, Kuhad R C, Lal B	102
Nirmala Agrawal, R.S. Sachan.....	102
Nirupama Mallick.....	103
Olivier Potin , Catherine Rafin, Etienne Veignie.	103
Omry Koren, Vishnia Knezevic, Eliora Z. Ron, Eugene Rosenberg	103
R. Bennicelli, Z. Stępniewska, A. Banach, K. Szajnocha, J. Ostrowski.	103
Riina Turpeinen, Timo Kairesalo, Max M. Häggblom.	104

Shardendu, N. Salhani, S. F. Boulyga, E. Stengel.	104
Shen-Yi Chen, Jih-Gaw Lin	105
Solange In [^] es Mussatto, In [^] es Conceição Roberto.	105
Sugijanto Kartosentono, Ana Nuraida, Gunawan Indrayanto, Noor Cholies Zaini.....	105
Thomas Pümpel, Lynne E. Macaskie, John A. Finlay, Ludo Diels, Marios Tsezos	106
W. M. Law, W. N. Lau, K. L. Lo, L. M. Wai, S. W. Chiu.....	106
Y. X. Chen, Q. Lin, Y. M. Luo, Y. F. He, S. J. Zhen, Y. L. Yu, G. M. Tian, M. H. Wong.	106
A. P. Soldatkin, J. Montoriol, W. Santc, C. Martelet, N. Jaffrezic-Renaulta.....	107
Anil K.	107
Caroline Michela, Fabienne Battaglia-Bruneta, Canh Tran Minhb, Mireille Bruschi and Ioannis Ignatiadis.....	107
Daniel Filippini, Tony P. M. Anderssonb, Samuel P. S. Svenssonb, Ingemar Lundströma..	108
Graham H. Crossa, Andrew A. Reevesa, Stuart Branda, Jonathan F. Popplewellb, Louise L. Peelb, Marcus J. Swannb, Neville J. Freeman.	108
Jiantao Pang, Chunhai Fan, Xinjian Liu, Ting Chen, Genxi Li.....	109
Kwang-Soup Song, Toshikatsu Sakaia, b, Hirofumi Kanazawaa, Yuta Arakia, Hitoshi Umezawaa, Minoru Tachikia, Hiroshi Kawaradaa.	109
M. Schwenk, T. Gabrio, O. Pöpke, T. Wallenhorst.	109
P. C. J. Roach, D. K. Ramsdena, J. Hughesb, P. Williamsa.	110
Seiji Shibasakia, Atsuo Tanakab, Mitsuyoshi Ueda..	110
Yang Liu, Xiao Yu, Rui Zhao, Di-Hua Shangguan, Zuyi Bo, Guoquan Liu.	110
Yuh-Ming Uanga, Tse-Chuan Chou.....	111
A. P. Mitra, C. Sharma..	111
Ingle ST, Pachpande B G, Mali D S.	112
Lal B, Mishra S, Bhattacharya D, Sarma P M.	112
Mohindru V K, Singh Kawaljeet, Jena B K, Siddiqui N A, Kulkarni A G.....	112
Sinha S and Sreekesh S.....	113
U. F. Wehmeier, W. Piepersberg.....	113
Bharat Bhushan, Louise Paquet,Jim C.Spain, Jalal Hawari.....	114
Bharat Bhushan, Sandra Trott, Jim C. Spain, Annamaria Halasz, Louise Paquet, Jalal Hawari.	114
Byung-Taek Oh, Patrick J. Shea, Rhae A. Drijber, Galina K. Vasilyeva, Gautam Sarath....	115
C. M. Kao, J. K. Liu, H. R. Lou, C. S. Lin, S. C. Chen.	115
Céline Tixier, Martine Sancelme, Selim Aït-Aïssa, Pascale Widehem, Frédérique Bonnemoy, Annie Cuer, Nicole Truffaut, Henri Veschambre.	116
D.Y. Bojinova, R.G. Velkova.....	116

Dai Kitamoto, Toru Ikegami, Gaby Tiemi Suzuki, Akira Sasaki, Yu-ichiro Takeyama, Yasushi Idemoto, Nobuyuki Koura, Hiroshi Yanagishita.....	117
David J. Hopper, Lisa Cottrell.....	117
Henry H. Tabak, Richard Scharp, John Burckle, Fred K. Kawahara, Rakesh Govind..	117
Inge S. Fomsgaard, Anne G. Mortensen, Sandra C. K. Carlsen..	118
Jörg Overhage, Alexander Steinbüchel, Horst Priefert	118
Kamlesh Kanwar, Anju Bhardwaj, Sandhya Agarwal & D R Sharma.	119
Keishiro Yoshida, Kazuo Furihata, Hiroshi Habe, Hisakazu Yamane, Toshio Omori.....	119
Koldo Aurrekoetxea-Hernández, Elena Buetti.	119
M. Bavcon, P. Trebše, L. Zupančič-Kralj.....	120
Manju Sharma, Rohini Sahni, Rekha Kansal, K R Koundal.....	120
María-Isabel Ramos-González, Arie Ben-Bassat, María-Jesús Campos, Juan L. Ramos.....	120
Michael Seeger, Myriam González, Beatriz Cámara, Liliana Muñoz, Emilio Ponce, Lorenzo Mejías, Carolina Mascayano, Yesseny Vásquez, Silvia Sepúlveda-Boza.	121
Mokashi S.A.,Paknikar K.M.	121
P. Pattanapitpaisal, N.L. Brown, L.E. Macaskie.....	122
Patil Y.B.; Paknikar K.M..	122
R. Schroll, F. Brahusi, U. Dörfler, S. Kühn, J. Fekete, J. C. Munch.....	122