ENHANCING CARBON SEQUESTRATION AND RECLAMATION OF DEGRADED LANDS WITH FOSSIL-FUEL COMBUSTION BYPRODUCTS

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INTRODUCTION

This joint ORNL and PNNL project examines the potential use of lands that have been disturbed by mining, highway construction, or poor management practices for terrestrial carbon sequestration. The approach includes examination of the effects of amendments with solid byproducts from fossil-fuel combustion, paper production, and biological waste-treatment facilities for enhancing carbon sequestration. The primary goal is to identify and quantify the key factors leading to successful C sequestration and reclamation of degraded lands and to communicate this information to the community. A scientific evaluation of existing field sites where amendments have been applied provides the basis for the guidelines. Results from the available literature are combined with additional new measurements of properties at these sites including (1) the extent and nature of the sequestered C, (2) microbial communities and their influence on greenhouse-gas emissions, and (3) redox, alkalinity, toxic metals, and key soil physical properties. Specific tasks focus on the accumulation of carbon in soil over time (including carbon sequestered in deep soil horizons) and the impact of environmental conditions on the rate of accumulation, assaying for populations of denitrifying bacteria that can effect emissions of greenhouse gases, assessing potential environmental health impacts (accomplished through a series of column leaching and toxicity experiments), comparing the results to DOE/CSiTE measurements, and evaluating management practices to optimize this sequestration strategy. Technology transfer workshops, presentations at academic and industrial meetings, and publications are employed to engage industry and create working partnerships. Project results will be summarized in a set of optimum site-management practices and practical guidelines, which include policy and technical considerations.

BACKGROUND

The concern for the potential global change consequences of increasing atmospheric CO_2 has necessitated the development of mechanisms to reduce or stabilize atmospheric CO_2. During the next several decades, a program focused on terrestrial sequestration processes can make a significant contribution to abating the increase, and easing the transition to other renewable energy sources. Within this context, the restoration of degraded soils represents an opportunity to couple carbon sequestration with the utilization of fossil fuel and energy byproducts and other waste material (e.g., industrial sludges, municipal biosolids, mulch) while achieving ecological and aesthetic benefits for society in improved soil quality, biodiversity, and ecosystem services.

The goal of this project is to study the use of fossil fuel byproducts to foster carbon sequestration in degraded lands. This has the triple benefits of carbon storage, byproduct utilization and land reclamation. This research will be conducted in conjunction with the DOE Center for Research on Enhancing Carbon Sequestration in Terrestrial Ecosystems (CSiTE), which will allow DOE to leverage existing CSiTE activities by expanding its research and partnerships to degraded lands and the use of fossil energy byproducts to stimulate carbon sequestration in those terrestrial ecosystems.

The potential of energy byproducts such as fly ash as soil amendments to enhance C sequestration in degraded lands can be most fully realized if these inorganic byproducts are applied in conjunction with organic amendments, including both mulch from biomass productivity, as well as process waste materials such as biosolids, and pulp and sludge from paper production. These organic amendments can have beneficial effects that complement and extend those of the inorganic fly ash material, including:

- Improving soil structure and moisture retention capacity of soil
- Releasing nutrient elements within the organic structure, adding pH buffering capacity, and acting as a metal-ion buffer in soil to make micronutrients available to plants.
- Stabilizing toxic metals in soil, thereby reducing their migration to groundwater and reducing their uptake and toxic effects in plants.

GOALS AND TECHNICAL APPROACH

The **goals of this project** encompass both scientific and technology transfer components:

Scientific Goal: Identify optimal selection and delivery strategies to maximize the contribution of amendments to carbon sequestration.

Technology Transfer Goal: Foster interactions between the scientific and user communities to maximize the application of new knowledge and approaches for enhancing terrestrial C sequestration through optimal utilization of fossil energy byproducts and management of degraded lands.

Our **research strategy** to achieve the Scientific Goal of optimizing C sequestration has three parts.

1. *Evaluate Existing Experimental Sites:* Because of the diversity and extent of previous amendment studies and the long times needed to determine success or failure from a C-sequestration perspective, we are evaluating a number of existing sites where fly ash and biosolids amendments have already been applied. These experimental sites were selected to represent different levels of amendments, different types of fly ash (alkaline and acid), and different ages since treatment. Data obtained from these experimental manipulations includes parameters such as the depth distribution and amounts and types of organic and inorganic C present, emission of greenhouse gases, and changes in nitrogen cycling microbial communities. This new information is being correlated with amendment treatment parameters and other soil properties to identify the most favorable (and detrimental) combinations of soils, fossil-fuel combustion byproducts, and management practices
2. *Conduct Laboratory Experiments to Identify Key Amendment Types and Potential Management Strategies:* A set of laboratory experiments to assess the relative impacts of different environmental conditions such as wetting and drying cycles, oxidizing and reducing conditions, and specific amendments that enhance the accumulation of C by soils are being conducted at PNNL. In addition, we are examining the potential for release of other greenhouse gases because of the amendments in greenhouse scale studies. This work will focus on nitrous oxide emissions as a result of nitrogen input via the amendments. The results of these bench-scale experiments will guide the design of field-scale experiments and help ensure their success.
3. *Conduct Laboratory Experiments to Assess the Environmental Implications of Coal Combustion Byproducts and Biosolid Amendment Utilization:* A series of experiments designed to address public concern over the release of toxic metals from fly ash and biosolid amendments will be accomplished through laboratory column leaching procedures. Results taken from this simulated weathering or leaching will be examined using a standard biosensor-based measurement technique for toxicity testing of water and soil.

Our current efforts to achieve the *Technology Transfer* goal are focused on communication but the focus will be expanding to include policy studies

1. *Communication*: The Regional Partnerships in Terrestrial Carbon Sequestration Workshop conference was sponsored by the U.S. Department of Energy and National Energy Technology Laboratory (NETL) and The DOE Consortium for Research on Enhancing Carbon Sequestration in Terrestrial Ecosystems (CSiTE) in November, 2001 in Lexington, KY. Presentations from the meeting and summaries of the breakout sessions can be found at: http://www.netl.doe.gov/publications/ proceedings/01/carbon seq terr/cseq-terr01.html. Recent presentations include, “Evaluation of

Microbial Communities in Soil Using a Mixed Functional and Phylogenic Array," by A. V. Palumbo, Z. Yang, S. M. Tiquia, L. Wu, R. A. Hurt, J. R. Tarver, L. S. Fisher, C. C. Brandt, and J.-Z. Zhou; and "Carbon Accumulation and Microbial Community Structure in Reclaimed Mine Soils," by S. M. Pfiffner, A. V. Palumbo, J. R. Tarver, L. S. Fisher, J. Cantu, and C. C. Brandt at the AGU Fall Meeting on December 6–10, 2002. "Development of Measurement Techniques for Carbon and Microbial Communities in Mine Soils," by A. V. Palumbo, M. Martin, Z. Yang, J. Tarver, S. Fisher, and S. Wullschleger was presented at the USDA Carbon Symposium on Natural Resource Management to Offset Greenhouse Gas Emissions in November, 2002. In addition, we plan on presenting the following presentations in 2003: "Issues with the Use of Fly Ash for Carbon Sequestration," by A. V. Palumbo, L. S. Fisher, J. R. Tarver, W. L. Daniels, Z. Yang, S. M. Tiquia, L. Wu, J.-Z. Zhou, and J. Amonette; and "Fly Ash Amendments Catalyze Soil Carbon Sequestration," by J. E. Amonette, J.-B. Kim, C. K. Russell, A. V. Palumbo, and W. L. Daniels at the 2nd Annual Conference on Carbon Sequestration sponsored by the Department of Energy and the National Energy Technology Laboratory (NETL) to be held May, 2003; "Microbial Communities, Carbon, and Nitrogen in Mine Soils Reclaimed with Fly Ash and Biosolid Amendments," by A. V. Palumbo, L. S. Fisher, J. R. Tarver, W. L. Daniels, Z. Yang, S. M. Tiquia, L. Wu, J.-Z. Zhou, and J. Amonette at the 2003 Annual ASM meeting on May 18–22; "Metals in Leachates of Soil, Biosolids, and Fly Ash," by A. V. Palumbo, L. S. Fisher, J. R. Tarver, J. E. Amonette, and W. L. Daniels, and "Enhancement of Soil Carbon Sequestration by Amendment with Fly Ash," by J. E. Amonette, J.-B. Kim, C. K. Russell, A. V. Palumbo, and W. L. Daniels at the 2003 Fly Ash Utilization Conference to be held October 20–23, 2003; and "Metal Leaching and Toxicity Measurements: Fly Ash Implications for Land Application," by S. Fisher, A. V. Palumbo, J. R. Tarver, J. E. Amonette, W. L. Daniels, and Y. Roh at the American Society of Agronomy Annual Meeting to be held on November 2–6, 2003. In addition, we have papers published (Martin et al. 2003), in press, and submitted.

2 *Policy Studies:* The potential effectiveness and efficacy of combining carbon sequestration and mine land reclamation has yet to be assessed from ecological, economic, social, and institutional perspectives. This approach seeks to undertake a portion of this assessment, by focusing on economic, social, and institutional issues that must be addressed for the technical information to be effectively applied. We envision a three-fold approach: (a) assessing the current situation in light of existing regulations and the values, goals, and positions taken by parties typically engaged in mine reclamation policy and decision making; (b) developing realistic scenarios to analyze the potential for carbon sequestration on reclaimed land; and (c) proposing realistic mechanisms for promoting carbon

sequestration on reclaimed land in ways that are likely to comply with regulations and have the potential to be acceptable (or, not highly objectionable) to involved constituency groups.

TECHNICAL PROGRESS

SOIL CARBON IN RECLAIMED MINE SOILS

Samples were taken from sites in the eastern United States, including West Virginia and Virginia (Lee Daniels) and from sites in the Midwest including several sites in Ohio (Rattan Lal). Samples were analyzed at Va. Tech, Ohio State, and ORNL. Results from this series of experiments were presented in the Fossil Energy 2001 Annual Report. In summary, mine soil C levels in samples taken from the Powell River Biosolids Project (PRS) PL experimental plots did not show a long-term effect of biosolid treatment on C levels in the surface soils (A horizon); however, deeper soil layers (AC horizon) do appear to be higher in organic C. While soil organic matter levels in the Controlled Placement mine soils showed a clear effect of the 1982 treatment, applications of biosolids and sawdust did not appear to differ in soil C levels from the untreated controls. Fly ash treatment also appeared to consistently increase soil C in the reclaimed mine soils.

MICROBIAL ANALYSIS

We examined the carbon content of reclaimed mine soils treated with soil amendments (e.g., fly ash and biosolids) for measures of soil diversity using terminal restriction fragment length polymorphism (TRFLP) and phospholipid fatty acid (PLFA) lipid biomarkers. We examined the diversity among fungal communities under various aspects of carbon content using TRFLP. TRFLP was run on 18S rDNA from polymerase chain reaction (PCR) amplification using primers specific for fungi. Results from the TRFLP (presented in last Annual Report) were compared to sequencing of 18S clones. Cloning and sequencing indicated that, along with the few dominant populations, there was a surprising diversity of fungal clones. Rarefraction analysis of the data indicated that although total diversity of fungal clones was greater than we were able to measure with this level of effort (Fig. 1) it appeared that we were able to effectively sample the dominant populations.

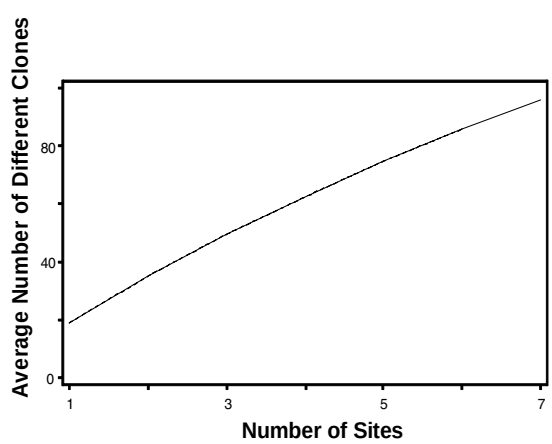

Fig. 1. Rarefraction analysis species area curve (including all sites).

The five most dominant populations (>50%) included *Mortierella verticillata*, a common acidophilous fungus in forest soils of temperate zones, *Sordaria fimicola*, a common coprophilous pyrnomycete found on fresh droppings of many herbivorous animals; *Mortierella chlamydospora*, generally found in agricultural fields and capable of decomposing chitin (Domsch et al. 1980); C*ryptococcus humicolus*, that has been isolated from acidic sandy soil supplied with benzene compounds and coke-plant wastewater (Middelhoven et al. 1992; Kwon et al. 2002); and *Cercophora septentrionalis*, which is often found on submerged wood (Domsch et al. 1980). The >50% pattern of sequences coming from only 5 clones is quite different from that observed in surface soils for bacteria where the most dominate populations account for just a few percent of the total (e.g., Zhou et al. 2002). However, Buchan et al. (2002) observed this dominance by a few cloned types for ascomycete fungi on Spartina alterniflora blades in a salt marsh.

Ordination techniques used to assess relationships among the sites in terms of fungal community structure revealed little consistency among samples from the same treatment (data not shown). One possible explanation is that, because cloning and sequencing techniques are of necessity done on only a small portion of the genetic material in the samples, the samples do not represent the community well. DNA microarrays could better assess the community (e.g., Wu et al. 2001) as these would assess 10^6 to 10^8 more genomes in each measurement than with cloning and sequencing. Thus, microarrays would sample a much larger portion of the population and could be applied to more sites.

We have used the clone data to design an array to be utilized on these sites. An array using many of the common species found in the clone libraries and a selection of relatively uncommon species was constructed and is being tested. The array also includes hundreds of sequences for bacterial functional genes. Thus, we hope to determine whether if the differences in carbon content related to the original treatments (e.g., fly ash and biosolids) are related to differences in the microbial community that can be assayed with this combined functional and phylogenetic array.

PLFA ANALYSIS

PLFA data indicated viable biomass for bacterial and fungal communities varied among amendment treatment, depth, and sites. The highest bacterial biomass was present in the Jenkins Farm A1 soil horizon containing the fly ash and sawdust amendment (Fig. 2). The total PLFA biomass ranged from 1.9 to 265 nmol/g, which corresponded to cell densities of 4.8×10^7 to 6.6×10^9 cells/g. It was also determined that biomass decreased with depth (A horizon: ≥120 noml/g, A2 horizon: 70–100 nmol/g, weak B horizon: 40–80 nmol/g).

Results from the principal component analysis (PCA) showed that low biomass samples (data from control S4 and mine soils) were influenced mostly by gram-positive-indicating PLFA biomarkers (Fig. 3).

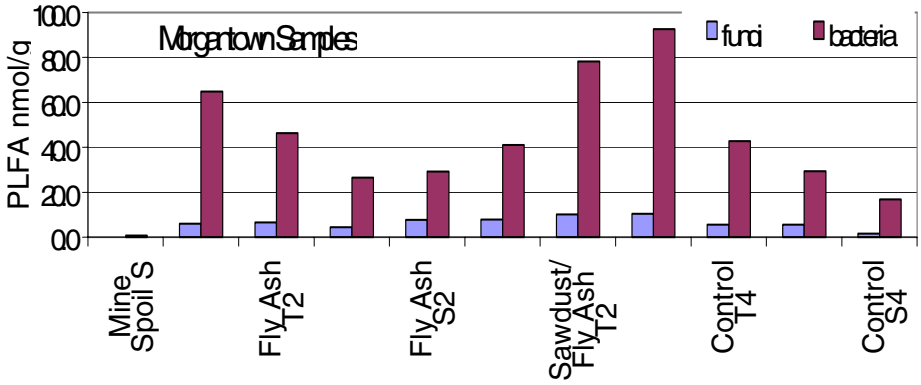

Fig. 2. PLFA biomass determined from surface mine spoils, fly ash treated plots (T2) Walls Farm 2 A1 horizon, (S2) Walls Farm 2 A1 horizon; sawdust and fly ash treated plots (T2) Jenkins Farm A1 horizon; and control sites (T4) Jenkins Farm A horizon and (S4) Jenkins Farm B horizon.

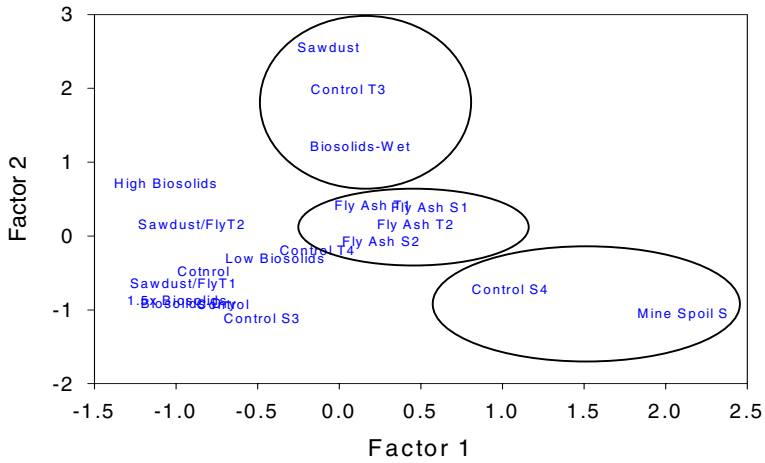

Fig. 3. Principle component analysis (PCA) performed using PLFA results from all treated plots.

Samples containing the highest amounts of carbon (fly ash treated T1, T2, S1, S2) were associated with PLFA biomarkers indicative of eukaryotes, especially those associated with plant residue (Fig. 3). These were determined to have been mostly samples taken from the plots amended with fly ash. PLFA generally indicative of gram-negative and eukaryotic microorganisms were found to influence samples collected from the plots amended with wet biosolid application and sawdust/fly ash material (Fig. 3).

FUNCTIONAL ANALYSIS

In addition to phylogenetic data, we are obtaining information about significant carbon cycling enzymes and metabolic processes. Twenty samples showed products using PCR amplification with

primers designed for lignase. We cloned the PCR products from 8 samples and have stared sequencing of the products. Additional genes will be targeted including those for tyrosinase (a fungal phenol oxidase) that is being used in other tasks on the project. Extracted DNA will also be hybridized in a microarray format to give information on the diversity and quantity of enzymes that are involved in aerobic metabolism, denitrification, iron reduction, sulfate reduction, and methanogenesis.

APPLICATION OF INNOVATIVE CARBON AND NITROGEN ANALYSIS METHODS

We have been testing the application of newer more rapid methods for carbon analysis to mine soils and have evaluated the use of laser-induced breakdown spectroscopy (LIBS) for determination of the total concentration of carbon and nitrogen in soils.

Our sampling generated a large range in soil carbon (less than 0.2 to approximately 8% in individual replicates) over which to generate and compare the LIBS technique to chemical oxidation and combustion methods (Walkley-Black and LECO). Soil carbon content measured by the LIBS technique exhibited a strong linear relationship for the previously mined soils when compared with the content measured by the LECO gas combustion technique (Fig. 4, insert) and the Walkley-Black method over the range of carbon up to approximately 6%. However, a sample at higher carbon (8.25%) did not fit the regression (Fig. 4, insert). Comparisons of the LECO technique and the Walkley-Black method appeared to have a nonlinear relationship at high amounts of carbon (Fig. 4). Apparently, there are discrepancies among the methods at high levels of carbon in soils.

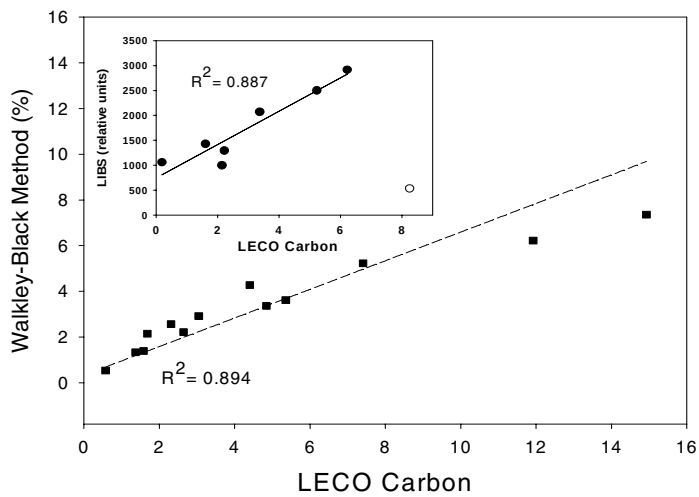

Fig. 4. Walkley-Black method compared to LECO gas combustion method for previously mined soils and LECO carbon calibration of LIBS data (insert). Discrepancies appear among the methods in these soils at above 6% carbon (as measured by LECO) where the LECO and Walkley-Black fit becomes non-linear (e.g., poor fit of regression at high carbon) and the LECO calibration fails (open circle on insert).

To achieve these results with the LIBS technique, it is advisable to make a large number of measurements. For example, although the range can be substantial, approaching 50% of the mean for some samples (e.g., Fig. 5), the standard error of the measurement is quite small (approximately 3%) when 20 measurements are made on a soil sample (Fig. 5). Thus, the major source of error is likely to be

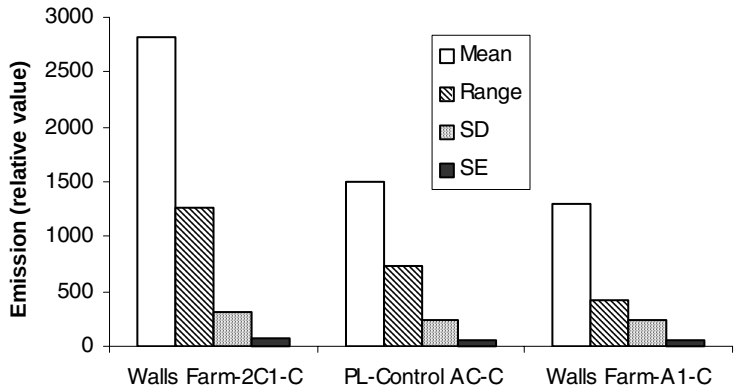

Fig. 5. LIBS measurements on twenty soil samples from previously mined soil.

sampling error, which must be addressed by taking multiple samples within a plot. A carbon assessment strategy should likely be based on application of the technique to a relatively large number of samples from a plot with a small number of subsamples (e.g., pellets).

POTENTIAL FOR LEACHING OF METALS

One significant concern in the utilization of coal combustion products is the potential for leaching of toxic metals from the fly ash. Our experiments were designed to examine the variability with regard to the source of the fly ash in leaching of metals under relatively mild extraction procedures that would be more relevant to environmental applications than standard EPA methods. We also designed experiments to determine if leaching of potentially toxic materials was influenced by mixing of the fly ash with soil and biosolids and if the biosolids could also be a concern for release of metals. We tested both type F and type C ash. Class F is produced from burning anthracite or bituminous coal and class C is produced from burning subbituminous coal and lignite. Class C also has cementitious properties and pozzolanic properties from the free lime (Stouraiti et al., 2002).

Both biosolids and fly ash appeared to contribute to leaching of some metals in the column leaching studies but for As and B only fly ash appeared to contribute to the leaching (Fig. 6). There were similar values in treatments receiving fly ash and biosolids for Mg, K, Ca, Fe, Ni, Cu, and Zn. The presence of elevated concentrations of these elements in the biosolid treatments that received fly ash indicated the contribution of the biosolids to the metals in the extracts. Elevated concentrations of Al and Na when fly ash was present with the biosolids clearly indicated the contribution of the fly ash to concentration of these elements in the effluent. As and B were found only in $CaCl_2$ extracts of samples treated with fly ash.

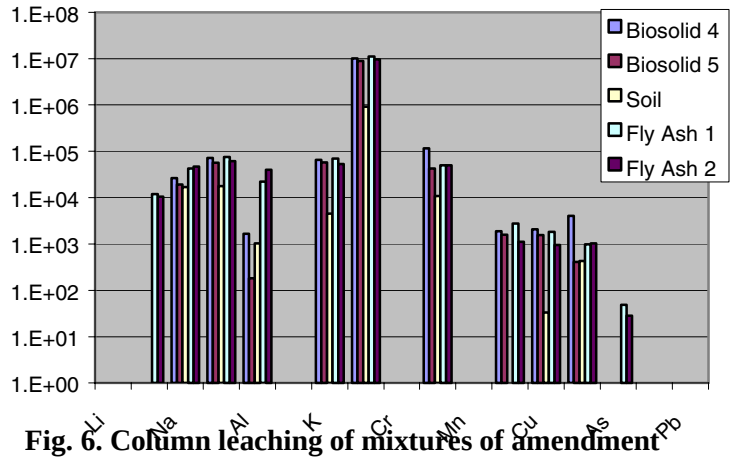

Fig. 6. Column leaching of mixtures of amendment materials. Ratios of fly ash/biosolids/soil are as follows: Biosolid 4 = 0/4/6; Biosolid 5 = 0/5/5; Soil = 0/0/10; Fly Ash 1 = 1/4/5; Fly Ash 2 = 2/3/5.

Differences in the presence and concentrations of the metals in the extracts between those from the column experiments (Fig. 6) and from the fly ash extractions (data not shown) may indicate the influence of the soil and biosolids in adsorption of specific elements. For example, Cr and Li were detectable in both hot water and acid extracts of the fly ash but was not detected in these experiments where the fly ash was combined with soil and biosolids. Although Si was detected in the hot water extracts of the fly ash it was not evident in the extracts from these experiments where the fly ash was combined with soil and biosolids. In addition, although Pb, and Cd were present in the acid extracts of the fly ash they also were not evident in the extracts from the columns with fly ash, soil, and biosolids.

Although potentially toxic metals can be leached from the fly ash using mild methods of leaching, for many of the most toxic elements leached the concentrations were very low. It was also evident from these studies that the TVA fly ash was considerably different that the western fly ash. The soil apparently reduced the leaching of several metals from the biosolids. Examination of combinations containing only soil and biosolids indicated that many of the metals detected in the analysis of the biosolids were not present in detectable amounts in the leachate of the column experiment. Although we detected As, Cr, and Cd in the biosolids none were detected in the column leaching experiment. Cu was detected in the biosolids and was leached at low levels from the biosolids treatments in the column experiments. Differences are likely due to the source of the coal, the post burning handling of the fly ash, or both. Leachates from TVA samples showed lower concentrations of some light elements and lower pHs.

Preliminary examinations of the data from measurements of toxicity using the Microtox system indicate the low levels of metals leaching do not result in detectable levels of toxicity. Experiments with a

variety of combinations of soils, fly ash, and biosolids give uniformly negative results for toxicity. Data analysis and experiments are continuing in this area.

LABORATORY ASSESSMENT OF POTENTIAL AMENDMENT TECHNOLOGIES

Research at PNNL seeks to understand the process by which organic-C is sequestered by soils (i.e., humification) and to develop practical approaches for enhancing the rate at which this sequestration occurs. Only a brief synopsis of that work is presented here. Experiments have focused on a model humification reaction involving a common phenoloxidase enzyme (tyrosinase) and organic monomers (polyphenols, hydroxybenzoic acids, and amino acids). A synergetic effect on the reaction rate when highly porous silica or manganese-oxide minerals are present in addition to the enzyme has been noted. As the porous silica bears some similarity to the cenospheres commonly found in fly ash, the effects of four alkaline fly ashes {Class C (sub-bituminous), Class F (bituminous), Class F [bituminous with flue-gas desulfurization (FGD) products], and Class F (lignitic)} on the reaction have been tested. The presence of FGD products completely halted the reaction, and the bituminous ash showed no benefit over an ash-free control. The sub-bituminous and lignitic fly ashes, however, increased the reaction rate by several-fold. In addition to their physical properties, the strong synergetic effect of these ashes may stem from their high Cu contents, as Cu activates tyrosinase, and from their alkaline pH.

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