Appendix A – CSiTE Accomplishments and 2005-2006 Progress Report*

Accomplishments

The overall CSiTE scientific approach has centered on integrated, hypothesis-driven science at multiple scales in the laboratory and the field. As illustrated in Figure 1, research progresses in an iterative manner with emerging data and scientific interpretation driving refinement of the scientific approach and feeding new information and knowledge to mechanistic and economic models. The models, in turn, inform assessments of sequestration capacity, technical feasibility, regional and national potential, and competitiveness with alternative greenhouse gas (GHG) mitigation technologies.

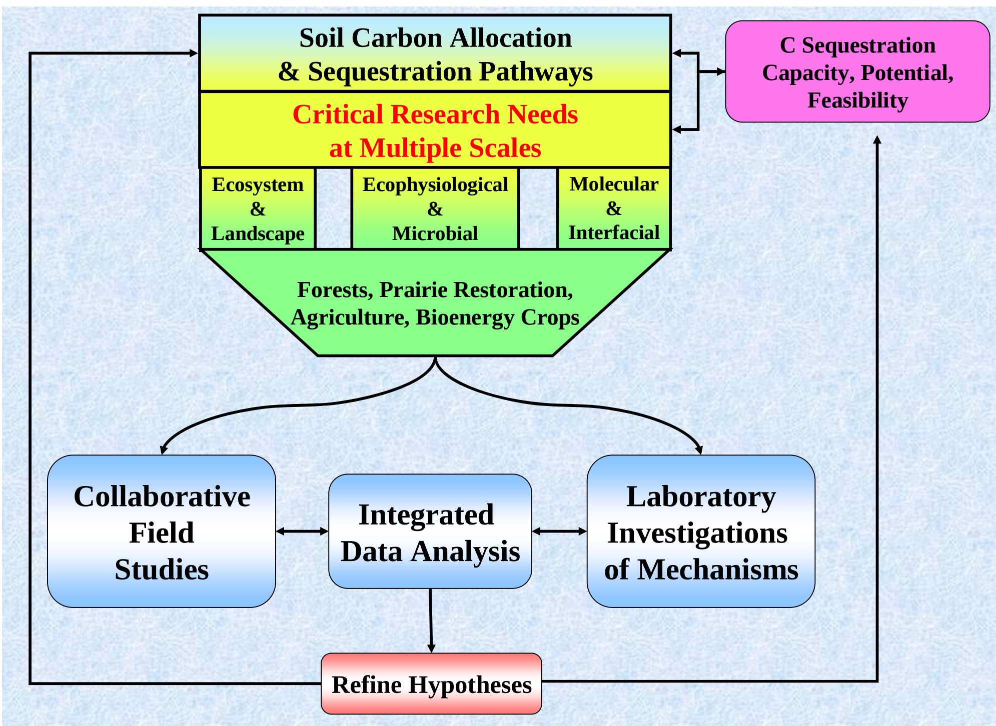

Figure 1. Conceptual model of CSiTE work flow and research output

A strategic decision was made when conceptualizing the CSiTE field campaign to identify "sites of opportunity" at which past land-use management decisions or changes over time (chronosequences) would act as surrogate experimental manipulations. The map in Figure 2 shows the locations of these sites, encompassing eastern hardwood and coniferous forest, western coniferous forest, various agricultural systems, grasslands and tallgrass prairie restoration. Results from 2 years of preliminary work at numerous sites was used to focus and integrate a more intense, CSiTE-wide program at four primary locations for the past 4 years: the Oak Ridge Reservation, the Fermilab prairie restoration site at Batavia, Illinois, the North Appalachian Agricultural Experiment Station at Coshocton, Ohio, and the Arid Lands Ecology

* Citations in this Appendix are included in Section V (Literature Cited) of the science plan.

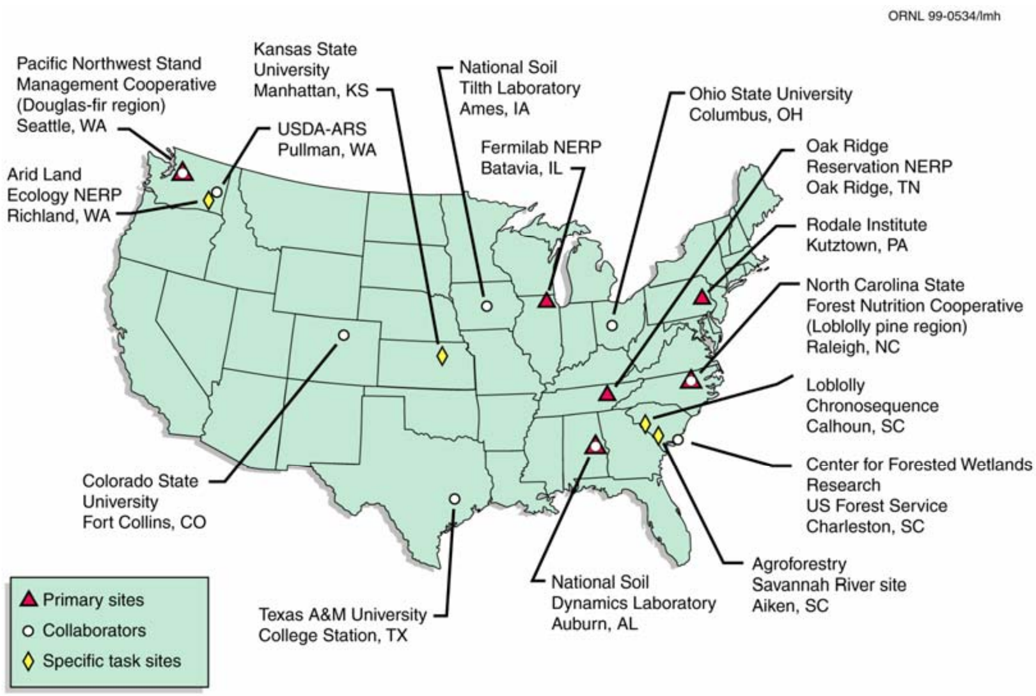

Figure 2. Map showing locations of original field sites and CSiTE collaborators.

Reserve at the Hanford site in eastern Washington state. Some smaller-scale projects were maintained as well with one example being DOE-NETL-sponsored research in collaboration with the USDA Forest Service at the Santee Experimental Forest at Charleston, South Carolina.

Figure 3 is a graphic representation of linkages among the original tasks showing how information from one task was dependent upon, supported another task, or both. As evidenced by more than 150 publications since 1999, our approach has resulted in greater research productivity than if the work been undertaken by individual principal investigators in isolation.

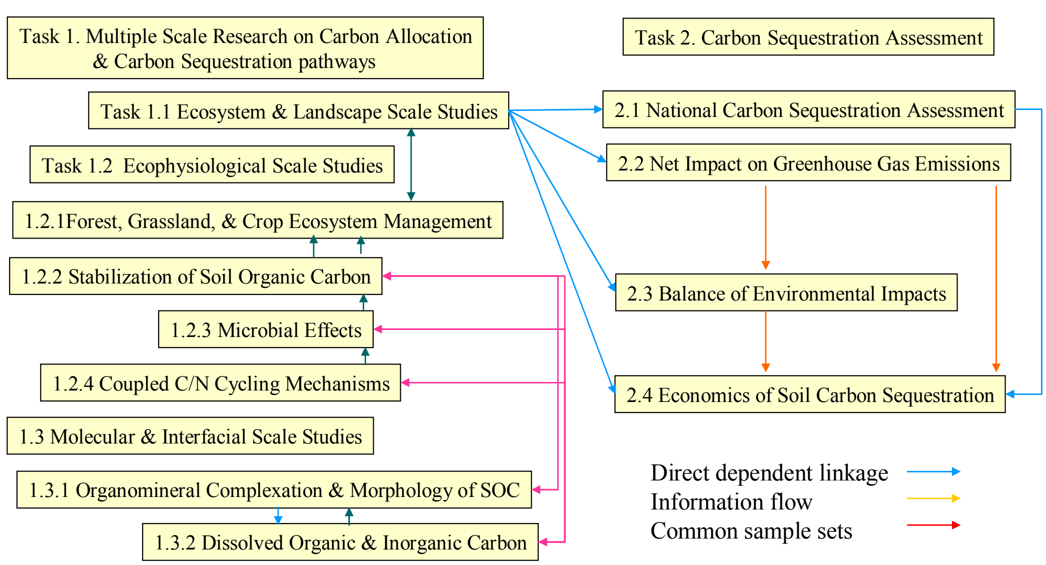

Figure 3. CSiTE original research tasks and task linkages.

At a higher level, CSiTE research has resulted in a number of significant accomplishments. These include:

Identification of manipulation concepts to enhance soil C sequestration. The seminal 2004 *BioScience* paper by Post et al., "Enhancement of carbon sequestration in U.S. soils," outlined a path from fundamental research to identification and implementation of novel concepts to enhance C sequestration. Following this approach, CSiTE researchers identified and are pursuing research to validate the feasibility of manipulation of soil physicochemical properties (e.g., pH, redox potential), urea for deep C storage, the use of soil amendments (e.g., fly ashes, black C) and land management strategies for enhancing soil C sequestration in a sustainable manner.

Elucidation of controls on the rates and limits of accumulation of soil organic matter. The development of new soil fractionation methods led to new insights on physicochemical controls over soil organic carbon (SOC) capture and longevity. Among other findings, the ability to partition particulate organic matter-silt-clay into non-aggregated, macroaggregated and microaggregated fractions resulted in demonstration that microaggregate protection increases longevity of clay-associated C and chemically resistant silt-associated C. Comparison of "old" vs. "new" C suggests that microaggregate fractions in 25 year prairie restoration experiments are not saturated. Mechanistic level understanding of humification chemistry has also been advanced.

Understanding the role of microorganisms in soil C processing at the community level. Methods were developed that improved fundamental understanding of the relative contribution to enhanced C sequestration of fungi and bacteria and the importance of mycorrhizae in long-term stabilization in minimally disturbed systems. Microarray technology for investigating microbial relationships to coupled C and N cycling processes was developed, resulting in the largest environmental nucleic acid array to date with over 20,000 genes.

Advances in modeling tools and their application to landscape-scale processes, full greenhouse gas (GHG) accounting, and economic assessments. CSiTE research has greatly improved our understanding of the environmental and economic consequences of land management practices to increase soil C sequestration. Enhancement and application of EPIC, APEX, and FASOM extended the basis for C accounting at the landscape level, comprehensive full GHG accounting, and incorporation of soil C in economic models for evaluation against other mitigation options. Transfer and utilization of components of the enhanced modeling tools has been made to U.S. EPA and the cross-agency CCTP.

2005-2006 Progress Report

The following progress report is organized by original task as proposed in 1999 and by which CSiTE has been organized and administered to date. Beginning in FY 2007, CSiTE will be re-organized by theme, as detailed in Section III.

Task 1. Carbon Allocation and Carbon Sequestration Pathways.

Ecosystem and landscape-scale understanding of soil C and N storage and dynamics are important to strategies for enhancing C sequestration in terrestrial ecosystems because land-cover management and land-use change present the best near-term options for enhancing soil C sequestration. The goal of Task 1.1 (Ecosystem and Landscape Scale Studies) has been to discover how land-use changes and complex land-management systems influence C and N cycling in vegetation and soil at the landscape scale. Results from observational and experimental field research in this task have and continue to be used to improve modeling efforts in Task 2.3 (described below) with the goal of achieving a quantitative understanding of soil C sequestration based on analysis of parameters such as plant growth, water balance, nutrient cycling and soil erosion. The role of litter inputs was studied in a forest system at the Oak Ridge Reservation while land management practices in prairie restoration and in different agricultural management systems were studied, respectively, at Fermilab and the North Appalachian Experimental Watershed in Coshocton, Ohio.

The goal of Task 1.2 (Ecophysiological Scale Studies) has been to extend fundamental understanding of microbial processes and soil aggregate properties that control soil C sequestration in managed and restored ecosystems. Principal research sites were Douglas fir stands in the Pacific Northwest and loblolly pine in the southeast, the Fermilab prairie restoration site, the Palouse of eastern Washington, and the Arid Lands Ecology Reserve at the Hanford site. Research was centered on forest nutrient management, grassland restoration from cropland, cropping systems, stabilization of SOC, microbial effects and coupled C/N cycling mechanisms. The goal of Task 1.3 (Molecular and Interfacial Scale Studies) has been to develop a basic understanding of fundamental interfacial and chemical processes that control formation of humus and organomineral complexes and how they govern dissolved organic and inorganic C storage and movement in soil. Research centered on laboratory- and pedon-scale field studies and included a regional-scale assessment of C sequestration potential in deep subsurface soils.

Forest Systems (1.1.1)

At Oak Ridge, significant topographic differences in soil C partitioning were found in the absence of detectable topographic differences in whole soil C stocks. Physical methods were used to partition whole soil C stocks into two pools with relatively fast (years) or slow (decades) turnover times. There was greater partitioning of C to the slow soil pool in mesic, N-rich valleys than on xeric, N-poor ridges and south-facing slopes. Topographic differences in N availability and soil C partitioning in site-specific studies followed the same patterns measured at the landscape scale. Measurements of annual leaf litter inputs, soil respiration, and C stocks in control plots were used to parameterize a two-compartment model of forest soil C dynamics.

Predicted fast and whole soil C stocks under leaf litter exclusion and supplemental leaf litter addition (triple ambient) were in good agreement ($r = 0.95$) with field measurements during the second year of the litter manipulation experiment. The turnover time of fast soil C at a valley site was approximately half that calculated for upland forests. Topographic differences in soil N availability did not translate to differences in forest soil C storage in a way that was easily detected using measurements of whole soil C stocks. At all sites, predicted soil C accrual over the short-term (decades) was primarily due to an accumulation of fast soil C. Both field measurements and modeling indicated that in a comparison of ridge, slope, and valley forests, mesic, N-rich valley soils are a more likely environment for long-term accumulation of soil C in the event of increased soil C inputs. A manuscript was prepared based on this research and submitted to *Water, Air, and Soil Pollution*.

Soil sampling at the litter manipulation experiment was conducted in 2001 and 2003. Simulations with a two-compartment model indicated that continuation of the experiment over a period of 6 years would produce a 6% loss and a 21 to 33% gain in whole soil C stocks, respectively, under the litter exclusion and the litter addition treatments at a ridge site. Predictions at the valley site over the same time period indicated an 11% loss in whole soil C stocks under litter exclusion and a 14 to 29% gain under supplemental leaf litter additions. At all study sites, predicted gains or losses in whole soil C could be attributed almost entirely to changing amounts of fast soil C. Annual leaf litter transfers or exclusions have been completed each year since the start of the experiment. A final soil sampling was conducted in May 2006 for the purpose of testing model predictions and the utility of the two-compartment soil C model. A manuscript (Topographic differences in forest soil C dynamics: implications for evaluating soil C sequestration potential) was submitted to *Water, Air, and Soil Pollution*.

Grassland Restoration from Cropland (1.1.2)

Plots in the Fermilab prairie chronosequence that were originally sampled in 1985 were resampled in 2004 to compare soil C stocks with samples from 1985, 1989, and 1999. Comparison of measured C values in 2004 to those predicted with a model based on 1985 measurements indicated that the space-for-time substitution of the chronosequence approach is reasonably accurate, although plot-level differences in rates of C accumulation were found. Carbon accrual rates in the surface 10 cm were sustained at linear rates over 19 years, with poorly drained prairies building carbon about 1.4 times faster than better-drained prairies. In contrast, a well-drained field planted with C3 Eurasian pasture grasses 3 years before the first prairie plot was restored has not gained carbon at a measurable rate since 1985. Measurement of stable isotopes at each time point indicated that C4-derived organic matter generally contributed more than C3-derived material to soil C accumulation. Although the rate of C accrual in restored prairies appears to be at least partly controlled by soil moisture, this study cannot resolve whether the difference in species composition (C3 Eurasian grass vs. the mixture of C4 and C3 species in the prairie) or differences in soil moisture or drainage conditions were responsible for the lack of C gain in the C3 grassland.

Research at the Fermilab chronosequence shows that restoration of prairie vegetation is highly effective at rebuilding SOC stocks at shallow depths at a rate of 0.33 to 1.5 Mg C ha^{-1} y^{-1}, depending on soil type. It also showed that cultivation of wet mesic soils causes a depletion of SOC at the depth of plowing but it results in a redistribution of carbon to deeper depths. In the remnant prairie, 77% of the total SOC (to a depth of 1 m) was present in the surface 25 cm of the soil profile. By comparison, only 64% or 68% of total SOC was found in the surface soils of cultivated land or the oldest restored prairies, respectively. Our results showed that restoration of tallgrass prairie can rapidly restore soil organic matter (SOM) lost through cultivation and has the potential to enhance SOC at depth. A manuscript was prepared based on this research and submitted to *Ecological Applications* (Matamala et al. 2006).

Studies also showed that C inputs to SOM are dominated by root and rhizome production. Within the first 12 years of restoration, the aboveground plant mass recovers to levels typical of a remnant prairie, but the recovery of the root system is slower and takes about 52 years. We have used the EPIC model with meteorological and edaphic data collected at Fermilab to simulate the annual rate of soil C accrual for the surface 15 cm of an agricultural field converted to a pure stand of big bluestem. The accrual rate was underestimated, mostly because the model predicted root and rhizome production inaccurately, particularly in the surface 5 cm. Other simulations are being produced to describe the mixture of C_3 and C_4 plants typical of the midwest tallgrass prairie ecosystem to simulate the particularities of the recovery of the vegetation and plant diversity to approximate rates of SOC accrual under restoration of native tallgrass vegetation.

A new experiment was conducted in the fall of 2005 to compare the distribution of SOC at depth in cultivated versus native lands. Five paired row crop-prairie remnant fields were sampled across Iowa, Illinois, and Wisconsin to represent wet-mesic soil types within the climate typical of the U.S. Corn Belt region. Each pair consisted on a historically known tallgrass prairie remnant and a nearby long-term-cultivated field, currently planted to corn, on the same soil type. The soils were sampled to 1 m depth, sectioned at intervals of 2.5 cm and 5 cm, and analyzed for SOC. The plots were compared by using a cumulative mass approach to account for variations in effective sampling depth and soil mass caused by cultivation-induced changes in bulk density. Preliminary results showed a decline in SOC in surface soils that accounted for a reduction of 12-32%. This decline was constrained to the depth of plowing, where decades of tillage have lowered and homogenized the concentration of carbon throughout the volume of soil mixed by tillage. However, we also found that total SOC to a depth of 1 m was significantly greater in cultivated compared to remnant soils. Preliminary data show that this increase varied from 8% to 30%, potentially accounting for as much as 4 kg C m^{-2}. At this time two hypotheses are being evaluated to explain these observations, either independently or working together:

(H1) Carbon gains at depth come from the mixing of surface and subsurface soil during tillage practices, which distributes soil with greater C concentrations deeper in the soil profile.

(H2) Carbon gains at depth are a result of increased downward transport of SOCs in percolating waters under cultivation.

These results suggest that the depletion of SOC in cultivated lands located on poorly drained soils in the U.S. Corn Belt has been largely overestimated. Thus, suggesting that it may be wrong to assume that the depletion of SOC at shallow depths represents past losses of C to the atmosphere. Rather, the redistribution of C to deeper soil profiles may actually enhance the sequestration potential of cultivated lands above the levels present under native vegetation, if the redistributed C can be maintained while surface concentrations are enhanced.

Cropping Systems (1.1.3)

The field experiments were conducted at the USDA North Appalachian Experimental Watershed (NAEW) in Coshocton County, Ohio. The NAEW research station was established in 1938 initially to study the effects of conventional and conservation management practices on soil erosion, runoff and water quality (Puget et al. 2005). The NAEW research station contains a series of small and large watersheds delineated by natural boundaries and artificial berms. These watersheds have historical records of environmental conditions, soil characteristics and distribution, crop productivity, management operations, and, in some cases, surface runoff and soil sediment yield. Puget et al. (2005) selected five distinctly managed watersheds to study the turnover rate and distribution of soil C in aggregate-size fractions and attempted to relate these to land use changes and soil management. The treatments selected were: 1) secondary forest (mixed white and red oaks with yellow poplars woodland), 2) meadow of orchard grass converted from no-till corn in 1988, 3) no-till (NT) continuous corn since 1970, 4) NT corn-soybean rotation with ryegrass as cover crop practiced since 1984, and 5) conventionally (moldboard) plowed (PT) continuous corn since 1984. In two other studies, Blanco-Canqui et al. (2005a,b) used seven long-term NAEW watersheds to study the influence of soil C content and management on soil strength and mechanical properties of soil aggregates. The seven treatments were categorized by degree of soil disturbance and use of organic amendments: 1) PT, chisel plow, disk + manure, NT + manure, NT, pasture, and forest.

Puget et al. (2005) observed large differences in SOC concentration among the treatments studied. In the top 5-cm depth, SOC concentration (g C kg^{-1}) was 44.0 in forest, 24.0 in meadow, 26.1 in NT corn, 19.5 in NT corn-soybean, and 11.1 in PT corn. The fraction of total C in corn residue converted to SOC was 12% for NT corn, 11% for NT corn-soybean, and 8% for PT corn. SOC concentration decreased with reduction in aggregate size while macro-aggregates contained 15-35% more SOC concentration than micro-aggregates. In comparison with SOC stocks under forest to 30-cm depth (64.6 Mg C ha^{-1}), the proportion of SOC depletion was 24.0% in meadow, 19.8% in NT corn, 26.8% in NT corn-soybean, and 35.1% in PT corn. SOC sequestration averaged 280 kg C ha^{-1} y^{-1} when converting from PT to NT practices.

Blanco-Canqui et al. (2005a) determined cone index (CI), shear strength, bulk density, volumetric water content, and SOC concentration were determined at the summit, backslope, and footslope landscape positions at various soil depths. In general, SOC concentration was slightly higher at footslope than at summit positions in the cultivated watersheds. Soil bulk density was lower at footslope than at summit in NT + manure (1.22 vs. 1.42 Mg m^{-3}) and chisel (1.34 vs. 1.47 Mg m^{-3}) treatments. The forest treatment had the lowest CI (0.19 MPa), shear strength (6.11 kPa), and soil bulk density (0.93 Mg m^{-3}) and the highest SOC concentration (62.7 g C kg^{-1}). The opposite was true for the PT treatment. The addition of manure decreased both CI and shear strength while it increased SOC concentration. Results showed that landscape positions had small effect on soil physical properties, but management, particularly the addition of manure, had large and significant effects on soil strength and SOC concentration. In complementary work, Blanco-Canqui (2005b) found that soil macro-aggregates had the lowest tensile strength and density of the same long-term treatments of the previous study. The addition of manure had a positive impact on soil aggregation while excessive tillage had a negative impact.

Stabilization of Soil Organic Carbon (1.2.2)

In a collaborative study with J. McCarthy at the University of Tennessee, processes underlying the sequestration of organic matter in soil microaggregates were studied at the submicron scale by using ultra-small-angle x-ray scattering (USAXS) at Argonne's Advanced Photon Source to evaluate the total porosity and organic-matter-filled porosity within microaggregates. The distribution of nano- and micropores (1 nm to 5 μm) in microaggregates was measured before and after removal of organic matter by combustion at 350°C. Long-term cultivated soils, restored prairies of increasing ages, and a remnant prairie at Fermilab exhibited differences in the proportion of organic-matter-filled pores. The dominant process affecting the accumulation of organic matter in microaggregates appeared to be protection in pores that became entirely filled with organic matter. The data suggest that physical protection of organic matter may occur via both spatial and kinetic limitations. The pool of organic matter in filled pores that is available to microbes may be restricted spatially to the small area at the throats of these pores. The efficiency of extracellular enzyme-mediated degradation may also be limited because of restricted diffusion of enzymes to organic matter inside filled pores. These barriers could also protect organic matter in pores large enough for microbes to enter if the large pores were "walled off" from microbes and their enzymes by an outer periphery of inaccessible pores filled with organic matter.

Microbial Effects (1.2.3) and Coupled C/N Cycling (1.2.4)

Cessation of agriculture and reconstruction of prairie at the Fermilab site increases total microbial biomass and increases the abundance of fungi, particularly arbuscular mycorrhizal fungi, relative to bacteria. We suggest 1) that this observation is caused primarily to reduced disturbance when tillage ceases, and 2) that early changes are reversed later in succession. Vegetation characters also appear to be important; high ratios of microbial cyclopropyl phospholipid to precursors indicate that gram-negative bacterial communities are under stress (i.e., in stationary growth) in agricultural but not prairie soils, probably because C inputs are low relative to N inputs. Although the strongest gradient is the response to cessation of agriculture, a secondary gradient related to successional time is more strongly tied to soil characters, particularly soil bulk density, SOC, and soil organic N. Although the ratio of fungi to bacteria increases with SOC in agricultural soils, this ratio decreases with SOC and with successional time in prairie soils. As a result, improved metabolic efficiency resulting from increased relative abundances of fungi is unlikely to be a mechanism enhancing C storage in these soils. Instead, we suggest that fungi contribute to C sequestration through their role in soil structure and inputs of recalcitrant compounds.

We also evaluated changes in soil microbial community structure with depth in the soil column and across the landscape, along a successional gradient of native prairie grassland restorations. We found that total microbial biomass declined strongly with depth and that the decline was largely attributable to changes in soil C, N, or both. Community composition shifted with depth and age; the relative abundance of sulfate-reducing bacteria increased with both depth and age, while gram-negative bacteria declined with depth. A large component of the depth-induced change in microbial community composition was undetermined, but it might be caused by anoxia lower in the soil column. By simultaneously examining shifts in microbial community structure in two dimensions (successional time and depth), we were able to decouple variables that are strongly correlated in surface soils and reveal indirect rather than direct impacts of soil C on microbial community composition in this system. The ratio of cyclopropyl phospholipids to their precursors increased to a depth of 50 cm and then declined. We suggest that this decline reflects changes in microbial species composition, rather than a decline in stress low in the soil column. We found similar patterns of change across the landscape, regardless of whether shallow soil or an integrated soil column was used in the analysis. This observation suggests that changes in composition of microbial communities across the landscape can be determined adequately from surface soils.

Sequestration Potential in Deep Subsurface Soils (1.3.1)

The area of each series in the STATSGO database was calculated. An area-weighted sample was selected from the list of STATSGO series and identified the most recently characterized pedons in the National Soil Survey Characterization (NSSC) database. The chemistry of the selected series was calculated for data for all series by great group to ensure that selected series were representative. Requests were made to USDA state and county agents to acquire samples from approximately 100 soil series from around the country to perform C sorption isotherms. Thus far, hundreds of horizons from 20 soil series have been obtained, and C sorption and soil characterization has been completed. Because of the magnitude of the request, a memorandum of understanding between DOE/ORNL and USDA was established, and cost estimates were solicited for obtaining additional soils. Because of the cost of obtaining additional soils, we have temporarily suspended additional requests for soils.

Results indicate that Alfisol, Ultisol, and Mollisol B horizons have good sorption capacity for sequestering organic C. This is likely because of their large clay content that is often coated with Fe-oxides. Their slightly to highly acidic pH condition also enhances the sequestration potential of these soils. Ultisols are extensive in the southeast and Alfisols are extensive in the Midwest. Both soil types are dominant east of the Mississippi River. Mollisols are dominant east of the Rocky Mountains and west of the Mississippi River. Thus, the decision was made to consolidate the effort and focus on these three soil orders. The non-trivial task of how to estimate missing bulk density data within the NSSC database was determined and published (Heuscher et al. 2005).

Manipulations to Enhance Subsurface Organic C Pools (1.3.2)

Our goal is to test and resolve the hypothesis that deep subsurface soils can accumulate organic C as a result of near surface manipulations. The effort involves the use of two highly instrumented in situ soil blocks on contrasting soil types and quantifies the impact of coupled hydrological, geochemical, and microbial processes on enhanced subsoil organic C sequestration.

In December 2004, and January 2005, shallow lysimeters were installed at depths of 5, 10, and 15 cm at both Melton and Walker Branch Soil Blocks. A litter pan lysimeter was also installed to monitor the C leaching in the Oa-Oe horizons of the soil. Samples were collected from the soil blocks after rain events and were brought back to the lab for analysis. As in previous years we ran the samples for C, volume data collected, precipitation, pH, bromide, chloride, sulfate, and nitrate. In April 2005, approximately 45 soil cores were taken from both soil blocks. The cores were taken at the same depths as the water sampling ports. These samples were given to Chuck Garten for CHN and $^{13}C/^{14}C$ analysis. This information would allow background information to be established prior to setting up a tracer study at the soil blocks in 2006.

Laboratory tracer studies were conducted on two intact soil cores were taken from the Walker Branch Soil Block area. The idea was to take two soil cores in an area near the soil block that would be representative of the soil in soil block area. Two intact cores were taken and brought back to the lab to be carved and placed into columns. Urea and bromide were used as tracers.

Data have been analyzed and allow us to determine controls for conducting the tracer study at the Melton and Walker Branch Soil Blocks in Summer 2006.

Storm driven transport of organic C through an Ultisol and Inceptisol suggested that both physical and geochemical processes control fate and transport of C through the soil profiles. The highly fractured Inceptisol exhibited the highest C flux during storm events, which is consistent with its more rapid flow and transport characteristics and lower organic C retention capacity relative to the Ultisol. Mesopore domains along dipping bedding planes served as conduits for organic C movement through the profile. Variability in organic C sorption was a function of solid0phase pH, indigenous sorbed organic C, and clay content. Both aromaticity and hydrophobicity measurements suggested that larger organic C molecules were being preferentially adsorbed by the solid phase during movement through the profile (Jardine et al. 2006). These results provide quantitative information on the significance of C credits in deep soil profiles.

Humification Chemistry (1.3.3)

Previous work in this task suggested that co-catalysis of humification occurs by three mechanisms involving physical stabilization of tyrosinase, direct oxidation of the monomers, and promotion of the oxidation and condensation steps by alkaline pH. Although tyrosinase activity is greatest at neutral pH, the large pH dependence of the condensation step drives the overall reaction to maximum rates under alkaline conditions. Following this hypothesis, liming of soils to slightly alkaline pH should enhance net C sequestration. Raising soil pH, however, also is likely to affect the activity of enzymes other than tyrosinase, such as various hydrolases. Hydrolase enzymes promote the breakdown of organic matter, and so the relevant question becomes one of whether the balance between humification and decomposition changes as the pH is altered. Preliminary evidence from the intermediate-scale experiment at the Santee suggests that the balance does change and that decomposition increases relative to humification as a result of raising the pH. As a consequence analytical capabilities were broadened to allow monitoring of a suite of enzymes including tyrosinase, peroxidase, phosphatase, sulfatase, and other hydrolases. In addition, raising pH tends to decrease sorption of DOC to soil surfaces and thereby promotes leaching of DOC into deeper portions of the soil profile. Some evidence for this effect was also observed in the Santee experiment, confirming that two possible “desequestration” mechanisms (hydrolysis and leaching) could occur as a result of raising soil pH by alkaline fly ash amendments.

Given the uncertain gain from the use of alkaline fly ash, and the beneficial results we observed for an amendment with a high-C ash in a calcareous soil, the research focus shifted our focus to the role of unburned C (including charcoal and high-C fly ash). A collaboration was initiated with The Energy Institute at Pennsylvania State University to supply four eastern fly ashes having high unburned C contents (as high as 50%) and moderate acidities (as opposed to alkalinity). The first round of experiments examined the sorption of tyrosinase enzyme to a collection of alkaline and acidic fly ashes, and the impact of this sorption on its activity. The results of these experiments clearly showed that unburned C has a strong sorption affinity for tyrosinase. A collaboration was also developed with the Eprida Corporation located in Athens, GA to test charcoal generated during their innovative hydrogen production process.