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## IDENTIFICATION OF POTENTIAL BACTERIAL SOURCES AND LEVELS, RED DUCK CREEK, MAYFIELD, KENTUCKY

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Our grant work was directed towards finding possible point sources for fecal contamination of Red Duck Creek in Mayfield, Kentucky. We chose this topic because Watershed Watch sampling results showed extremely high e-coli colonies at various locations along the creek. From the Watershed Watch data, we believed a single pollutant source could be isolated.

For the data collection, we chose 12 spots scattered throughout the residential and commercial area of Mayfield. The locations were chosen based on possible contamination sources. We defined possible contamination sources as being sewage pipes, run off from agriculture areas, run off from industry, and possibly, unidentified septic systems. At each location we measured dissolved oxygen, turbidity, conductivity, temperature, pH, total coliforms, and e-coli colonies. Flows were generally too low to measure. Red Duck Creek rapidly recedes following precipitation and is typically ponded rather than flowing during low flow conditions.

The results from our data turned out to be inconclusive. Although some sampling events showed high levels of coliforms, we had no clear trend at the various locations. The overall results were uniform throughout the stream. Results from up stream and down stream of Mayfield could not be statistically differentiated. With no clear statistical evidence to point to any specific location, specific sources could not be determined.

Since specific sources could not be identified, widely distributed non-point runoff may have been the largest contributor for contamination of Red Duck Creek. Contamination sources could have included animal feces since domestic pets are numerous around Red Duck Creek, feces from birds that fly over and rest in nearby trees and in the water, and domestic livestock upstream of Mayfield.

Due to a funding delay, we were only able to sample in the Fall rather than Spring and Fall as intended. Hence, high flow data could not be obtained. Also, some local industries are seasonal and operate at a reduced level in the Fall. Furthermore, inputs from sewer lines and septic systems may have been reduced due to less infiltration and inflow. Additional research might include sampling in wetter seasons and running DNA tests on the bacteria to better differentiate possible sources.

## NOTES

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## WATER-QUALITY TREND ANALYSIS FOR STREAMS IN KENTUCKY

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Increasingly complex water-management decisions require water-quality monitoring programs that provide data for multiple purposes, including trend analyses to detect if water quality has improved, deteriorated, or remained stable with time. Identifying reasons for changes in water quality (natural or anthropogenic causes) is an additional component of trend analyses.

Trends in surface-water quality for 16 properties and water-quality constituents were analyzed at 37 sites with drainage basins ranging in size from 62 mi<sup>2</sup> to 6,431 mi<sup>2</sup>. Analyses of nutrients, major ions (chloride, sulfate), suspended solids, select metals (iron, manganese), fecal coliform, and select physical properties (pH, temperature, dissolved oxygen, specific conductance, and hardness) were compiled from the State's ambient water-quality monitoring network. Trends analyses were completed using the S-Plus statistical software program S-Estimate Trend (S-ESTREND) that detects trends in water-quality data. The trend detection techniques supplied by this software include the Seasonal Kendall nonparametric methods for use with uncensored data or data censored with only one reporting limit (Hirsch and others, 1982), and the Tobit parametric method for use with data censored with multiple reporting limits (Cohn, 1988). One of these tests was selected for each property and water-quality constituent and applied to all site records so results of the trend procedure could be compared among sites.

Statewide results indicate scattered statistically significant (p-value <0.05) upward or downward trends for most water-quality constituents or properties. A summary of selected results for nutrients, total suspended solids, chloride and sulfate is shown for 31 of the 37 sites in table 1. The 6 remaining sites had not been analyzed at the time of the abstract preparation.

### REFERENCES CITED

- Hirsch, R.M., Slack, J.R., and Smith, R.A., 1982, Techniques of trend analysis for monthly water quality data: *Water Resources Research*, v.18, no.1, p. 107-121.
- Cohn, T.A., 1988, Adjusted maximum likelihood estimation of the moments of lognormal populations from type 1 censored samples: U.S. Geological Survey Open-File Report 88-350, 34 p.

**Table 1. Summary of selected trend results for nutrients, total suspended solids, chloride, and sulfate at selected Kentucky Division of Water ambient water-quality network sites.**

[WV, West Virginia, TN, total nitrogen, NO<sub>3</sub>, nitrate; TKN, total Kjeldahl nitrogen; TP, total phosphorus; TSS, total suspended solids; Cl, chloride; SO<sub>4</sub>, sulfate; +, increased trend; --, decreased trend; blank space, no significant trend]

| Site name                                 | Site number | Trends |                 |     |    |     |    |                 |
|---|-------------|--------|-----------------|-----|----|-----|----|-----------------|
|   |             | TN     | NO <sub>3</sub> | TKN | TP | TSS | Cl | SO <sub>4</sub> |
| Tug Fork at Kermit, WV                    | PRI002      |        |                 | --  | -- |     |    | +               |
| Levisa Fork near Pikeville                | PRI006      |        |                 |     | -- | --  | +  |                 |
| Cumberland River near Burkesville         | PRI007      | --     | --              | --  | -- | --  |    |                 |
| South Fork Cumberland River at Blue Heron | PRI008      | --     | --              |     | -- | --  | -- | --              |
| Rockcastle River at Billows               | PRI010      | --     |                 |     | -- | --  |    | --              |
| Pond River near Sacramento                | PRI012      |        |                 |     |    |     |    |                 |
| Rough River near Dundee                   | PRI014      | --     | --              | --  | -- |     |    | --              |
| Green River at Munfordville               | PRI018      |        |                 | --  | -- |     |    |                 |
| Nolin River at White Mills                | PRI021      |        | +               | --  |    | --  | +  |                 |
| Eagle Creek at Glencoe                    | PRI022      | --     |                 | --  |    | --  | +  |                 |
| Kentucky River at Lock 4                  | PRI024      | --     |                 | --  |    |     | -- |                 |
| Salt River at Shephardsville              | PRI029      | --     | --              | --  | -- |     |    |                 |
| North Fork Kentucky River at Jackson      | PRI031      |        |                 | --  | -- |     | +  |                 |
| Middle Fork Kentucky River at Tallega     | PRI032      |        |                 |     | +  |     |    | +               |
| South Fork Kentucky River at Booneville   | PRI033      | --     |                 |     |    |     |    |                 |
| Beech Fork near Maud                      | PRI041      |        |                 | --  | -- | --  | +  |                 |
| Little River near Cadiz                   | PRI043      | +      |                 | --  | -- |     | +  |                 |
| Dix River near Danville                   | PRI045      |        |                 | --  | -- |     | +  |                 |
| Red River at Clay City                    | PRI046      |        |                 | --  | +  |     |    |                 |
| Little Sandy at Argillite                 | PRI049      | +      | +               | --  |    | --  |    | +               |
| Horse Lick Creek near Lamero              | PRI051      |        | --              |     |    | --  |    | --              |
| Salt River at Glensboro                   | PRI052      |        |                 |     | -- |     | +  |                 |
| Green River at Livermore                  | PRI055      |        |                 |     |    |     |    |                 |
| Mud River near Gus                        | PRI056      |        |                 |     | -- |     |    | --              |
| Rolling Fork near Lebanon Junction        | PRI057      |        |                 |     |    |     | +  |                 |
| Kentucky River near Trapp                 | PRI058      |        |                 |     |    |     |    | +               |
| North Fork Licking River near Milford     | PRI060      |        |                 |     | -- |     | +  |                 |
| Licking River at Claysville               | PRI061      |        |                 |     |    |     | +  |                 |
| Levisa Fork near Louisa                   | PRI064      |        |                 |     |    |     | +  | +               |
| Kentucky River at Lockport                | PRI066      |        | +               | --  | +  |     |    | +               |
| Kentucky River at High Bridge             | PRI067      |        |                 |     |    | +   |    |                 |

PRELIMINARY RESULTS OF A FECAL MICROBE SURVEY IN AN EUTROPHIC LAKE, WILGREEN LAKE, MADISON COUNTY, KENTUCKY

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Wilgreen Lake is a small (~14 mi<sup>2</sup>), eutrophic lake formed by damming several tributary streams to Silver Creek, Madison County, Kentucky. The lake receives runoff from industrial and urban areas (Richmond) that comprise ~10% of the total watershed area; most runoff is from cattle pasture or human developments encircling the lake. Present and past developments are on septic systems, and effluent from these systems is known qualitatively to seep into lake waters.

Our research group is currently conducting a study of the lake in order to identify major nutrient sources, and one possible tracer method is to quantitatively assay species-specific microbes in lake waters. In preparation for this effort in the 2007 field season, we sampled lake waters in July and August 2006 to characterize the spatial distribution and abundance of fecal microbes. Sampling stations (15 in number) encompass the lake's breadth and include samples from not only the trunk of the lake system (where deeper water occurs) but also from 3 tributaries – two of which have possible inputs from septic systems. We use the *Colisure* method from IDEXX Laboratories to determine the most probable number (MPN) of total coliform and *Escherichia coli* bacteria.

Higher numbers of fecal microbes occur in the two most densely populated tributaries, and we note 14 cases (at 8 sites) where assays exceed maximum standards of the EPA for bathing exposure (200 cfu per 100 mL for total coliform, TC; 235 cfu per 100 mL for *E. coli*, *EC*). The trunk locations show low numbers of fecal microbes (TC generally <150 cfu per 100 mL; *EC* generally <20 cfu per 50 mL) whereas the upper reaches of both Taylor's Fork and Old Town Branch show higher microbial abundance (TC generally >300 cfu per 100 mL; *EC* cfu generally >100 per 100 mL). Another tributary stream with no apparent human effluent at present shows much lower fecal microbe abundance. From the data, we infer there is significant input from septic systems into these specific regions of the lake. There are several other sources that must be eliminated as possibilities, but it is likely that the source of these fecal microbes is from septic systems encircling the lake. Substantial residential development is underway around Wilgreen Lake at present, and we intend that 2007 field results inform development practices.

*Kentucky Water Resources Research Institute Symposium, March 2007.*

## **NOTES**

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PRELIMINARY PHYSICAL AND CHEMICAL CHARACTERISTICS OF AN  
EUTROPHIC LAKE, WILGREEN LAKE, MADISON COUNTY, KENTUCKY

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Wilgreen Lake (Madison County, Kentucky) has been listed as “nutrient impaired” by the State, thus identifying the sources of nutrient (nitrogen and phosphorus) input into the system is critical in establishing best-practices management of this watershed. The lake’s drainage basin is diverse, receiving water from urban and industrial areas, residential property, and pasture land. Prior to tracer studies that will commence in the 2007 field season, we gathered framework data necessary to understanding the physical, chemical, and biological characteristics of Wilgreen Lake.

We established 9 water sampling stations and 16 data stations over the expanse of the lake, including its tributary and trunk drainages. Maximum water depth is about 16 meters near the dam. At all these stations we use an YSI probe to take temperature, conductivity, pH, and oxygen concentration measurements at depth intervals of 1 meter. We also collect water samples at depth intervals of 1 meter at appropriate stations. The chemical data we report here include ammonium ( $\text{NH}_4^+$ ) nitrogen concentration measured by the colorimetric method of Solorzano (1969). We gathered data monthly from June through September, taking probe data an additional time during September.

Preliminary data show that Wilgreen Lake is a typical eutrophic system. The lake was already stratified with our first sampling (31 May), showing a strong thermocline between 2 and 7 meters. Thus, the lake is segregated through the summer into a top and bottom layer, each with stark differences in their physical and chemical properties. Conductivity values in the upper layer are  $0.4 \pm 0.05 \mu\text{S}/\text{cm}^2$  and increase in along the thermocline reaching maximum values ( $\geq 0.55 \mu\text{S}/\text{cm}^2$ ) near the bottom. The top layer is alkaline (pH 8 - 9), whereas bottom waters are more acidic (pH 7 - 7.5). Dissolved oxygen concentration is highest in the top layer (8 -13 mL/L) with disoxic (0 - 2 mL/L) and anoxic (0 mL/L) waters in the bottom layer. Ammonium concentration is near zero in the upper layer and increases with depth in the lower layer to 5 ppm. Over the summer, the disoxic-oxic boundary remains at a depth of 3 to 4 meters, but anoxia moves upward in the water column, sharpening the oxygen gradient. The lake is surprisingly homogeneous, both physically and chemically, at the deeper-water sites. Shallow sites at the inlets are more variable. In some cases, surface waters there contain markedly less dissolved oxygen, perhaps due to higher oxygen demand and/or decreased photosynthesis.

*Kentucky Water Resources Research Institute Symposium, March 2007.*



## NOTES

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## LETHAL AND SUBLETHAL EFFECTS OF NUTRIENT POLLUTION ON AMPHIBIANS

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Fertilizer application is a widespread practice that greatly affects aquatic ecosystems in a variety of ways. One important affect is the toxicity of nutrients to various species of amphibians. Previous studies show that nitrate toxicity could be a major problem for amphibians at elevated levels, though few studies examine sublethal effects. On the other hand, phosphate has not been evaluated at all for toxic effects. To examine nutrient toxicity, laboratory experiments were conducted to measure the response of American Toad and Cope's Gray Treefrog tadpoles to various concentrations of nitrate and phosphate. The phosphate experiments exposed Cope's Gray Treefrogs to five treatments with concentrations ranging from 0 to 200 mg/L P-PO<sub>4</sub> and lasted for 15 days. Nitrate was examined in both the American Toad and Cope's Gray Treefrog using seven treatments: four with constant concentrations, ranging from 0 to 5 mg/L N-NO<sub>3</sub>, and three pulses that simulated the quick increase in concentration to 5 mg/L and slow decline that would be associated with runoff from a rain event. Pulses were timed at different points during development to determine critical developmental stages when tadpoles are most vulnerable. The amphibian responses that were measured included mortality, body condition, size at metamorphosis, time to metamorphosis, fluctuating asymmetry, directional asymmetry, and deformities. Phosphate was found to have no effect on any of the lethal or sublethal responses in Cope's Gray Treefrogs, indicating that phosphate may not be toxic to this species. Nitrate had no effect on American Toads but did affect the Cope's Gray Treefrog. Individuals from the treatment with the pulse late in development had more extreme directional asymmetry than other treatments in calcaneum length. The late pulse occurred during hind limb development; directional asymmetry in a hind limb trait suggests the disruption of the developmental process during the sudden increase in the concentration of nitrate. Individuals from the early and middle pulses may have had similar disruptions in development but were able to compensate prior to metamorphosis.



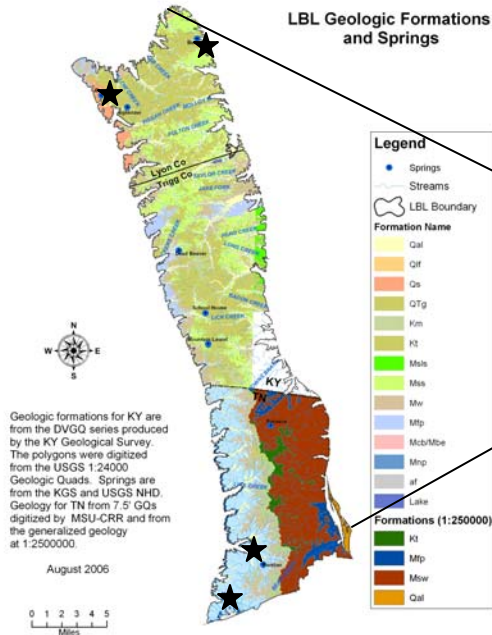
DIATOM COLONIZATION PATTERNS IN SPRINGS AT LAND-BETWEEN-THE- LAKES NATIONAL RECREATION AREA,  
WESTERN KENTUCKY AND TENNESSEE

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Diatoms (Bacillariophyta) are algae known for their sensitivity to chemical conditions in water. Therefore, they are a useful supplement to chemical analyses in assessments of water quality. Parent geology determines conductivity, alkalinity, pH and nutrient concentrations in spring ground waters that ultimately seep into streams. Spring water chemical factors may influence species composition of periphyton colonizing stream substrates.

In October 2006, an exploratory study of diatom colonization patterns was carried out in several springs emerging from different geological materials in Land-Between-the Lakes National Recreation Area located in western Kentucky and Tennessee (Figure 1). Two springs emerge from limestone geology (Panther and Mint in the south) and two springs emerge from siliceous/argillaceous geology (Barnett and Brown in the north). Unglazed quarry tiles were deployed in each stream a few meters downstream from each spring and were allowed to colonize for four weeks. The tiles were retrieved, diatoms were identified to genus, and biomass was determined from chlorophyll *a* analysis.

Physiochemical characteristics and nutrient concentrations also were measured in each spring.



There were significant differences between at least two of the springs in dissolved oxygen, alkalinity, pH, DO, turbidity, SiO<sub>2</sub>, SO<sub>4</sub>, Cl, and SRP concentrations. Discharge, chl *a*, NO<sub>3</sub>+NO<sub>2</sub> and NH<sub>4</sub> were not significantly different among any of the springs. Only conductivity was significantly different among all four

springs. *Achnanthes*, *Cocconeis* and *Gomphonema* were dominant in the carbonate streams with limestone geology (Panther and Mint springs); these taxa are calciphilous and high conductivity (107–481 uS cm<sup>-1</sup>) provide optimal conditions for growth. *Diatoma*, *Eunotia*, and *Pinnularia* were more abundant in the streams with siliceous and argillaceous geology (Brown and Barrett springs); these taxa are considered acidophilous and low conductivities (48–163 uS cm<sup>-1</sup>) are optimal for growth.

This study will be expanded over the next 8 months to test the hypothesis that springs with contrasting geologies (and therefore different water chemistries) will develop different diatom community compositions. Developmental sequence analysis of stream periphyton also will be used to test the hypothesis that different spring water origins as defined by stable isotope (<sup>13</sup>C) signatures will be reflected in the biomass of periphyton colonizing the substrata over time.

# DENITRIFIER ECOLOGY IN FRAGIPAN SOILS OF KENTUCKY

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Fragipans provide conditions conducive to denitrification because of seasonally perched water. Previous work on denitrifiers in fragipan soils indicated that surface manure addition could increase denitrifiers at all soil depths and cause stratification above fragipans (Fig. 1)(Fairchild et al., 1999). However, evaluating denitrifier populations by culture alone is not comprehensive because of the limitations of culture-based techniques. Functional genes involved in denitrification have been extensively and effectively used to study the highly diverse denitrifying bacteria in various soil and marine environments (Braker et al., 1998). In particular, the structural genes for Cu- (NirK) and heme- (NirS) nitrite reductases and nitrous oxide reductase (NosZ) have been used to characterize denitrifier communities. However, this has not extended to the fragipan/soil interface.

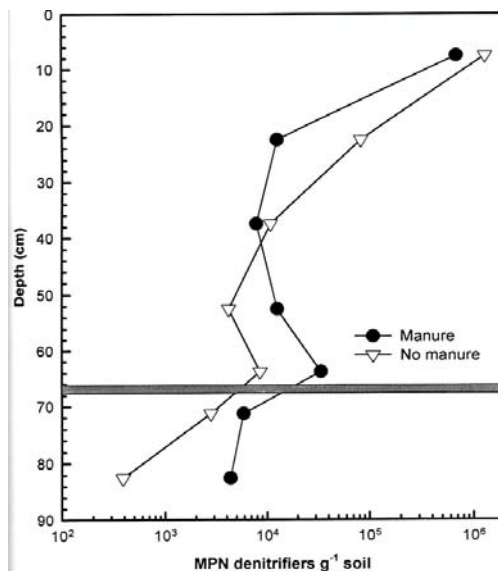


Figure 1. Manure effects on stratification of denitrifiers (Fairchild et al., 1999).

In our study we compared the denitrifier composition between surface and subsurface boundary layer soils from Princeton, KY in a cultivated site previously amended with poultry litter, and an unamended pasture site - both with identified fragipans. Most probable number (MPN) and denitrification enzyme assay (DEA) were used to determine the denitrifier population and activities, respectively. DNA based molecular analysis was used to compare denitrifier populations from soil of different depths.

The MPN results showed that denitrifier populations decreased exponentially from the soil surface to the depth of fragipan, and were stratified above the fragipan in some samples at both sites (Fig. 2). Nutrient analysis of soils from the cultivated site showed no significant difference in total C and N at the similar depths

between the plots with and without previous poultry litter amendment. Correspondingly, neither was there significant difference in denitrifier population size.

The DEA results showed higher denitrifier activity in the surface soil than in the boundary layer soils, which corresponded with our MPN data. However, due to the low

denitrifier populations at the fragipan layers, denitrifier activity in soil immediately above or below the fragipans was minimal at both sites.

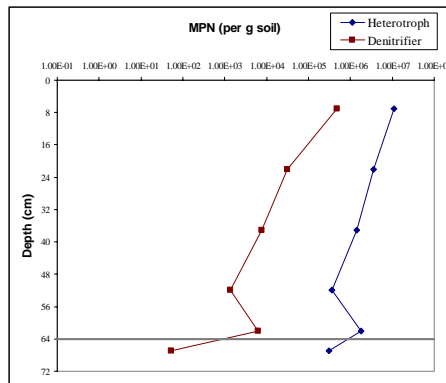


Figure 2. Denitrifier populations vs. depth in a pasture soil with fragipan appearing at 62cm.

The band densities of extracted DNA from soils at each site and depth indicated a dramatic decrease in the microbial population with the increasing depth. DNA recovery could be improved by adding skim milk to the commercial DNA extraction kits. Primers for *nirS* and *nirK*, encoding nitrite reductase, which catalyzes the rate-limiting step in denitrification, were used for the PCR amplification of DNA extracted from the various soils by depth. The Cu-type nitrite reductase NirK was found in both surface and subsurface soils while heme-type NirS enzyme was only found in the surface samples. These results suggest that either different denitrifier community structures occurred at different depth of soils, or the published primer sequences for *nirS* were less effective at reflecting the denitrifier populations existing in the fragipan soil environment.

The preliminary data from MPN and DEA analysis suggest that perched water alone is insufficient to drive significant denitrifier population changes at the fragipan boundary layer. While results of the nitrite reductase gene analysis suggest that the community structure of denitrifiers differs by depth in these soil environments, and potentially could contain unique types of denitrifiers. Our future study will focus on attempting to quantify changes in the denitrifier community structure and to manipulate denitrifiers at the fragipan boundary layers by surface amendments such as additional C and N.

## References

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LEVERAGING PARTNERSHIPS FOR IMPROVED RESEARCH TRANSLATION:  
THE UNIVERSITY OF KENTUCKY  
SUPERFUND BASIC RESEARCH PROGRAM  
RESEARCH TRANSLATION AND COMMUNITY OUTREACH CORES

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The University of Kentucky Superfund Basic Research Program Research Translation (RT) and Community Outreach Cores (COC) have forged a close relationship to promote improved communication with a number of audiences, including government agencies, impacted populations, and industry and healthcare professionals. Working together, the RT and COC have created an operational model that allows for optimal division of communication labor while promoting audience feedback for quantifiable measures of success. Among the University of Kentucky's ongoing RT and COC efforts are Superfund Community Action through Nutrition programs in impacted communities, the creation and maintenance of an RT database, the hosting of both academic workshops and public forums, and the development of an innovative tailored website. In addition, the UK-SBRP RT and COC leverage partnerships with a number of stakeholders to ensure successful, widespread communication of research findings and their implications for specific constituencies.

Supported by NIEHS/NIH (P42ES07380)





## EFFECT OF WASTEWATER TREATMENT PLANT EFFLUENT ON AQUATIC MICROBIAL COMMUNITIES

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Little is known about the effect of wastewater on stream microbial communities. Water samples were taken upstream and downstream from the wastewater treatment plant on Tates Creek which runs from Richmond, Kentucky to the Kentucky River. At the time of sampling, Hach water testing kits were used to measure chloride, dissolved oxygen, ammonium, nitrate, and phosphate. Nutrient increases were associated with wastewater effluent addition to the creek. Total heterotrophic counts were performed using R2A agar. Bacterial numbers peaked one mile down stream from the wastewater treatment plant but the number then decreased to pre-treatment levels further down stream. Microbial community structure was assessed using Biolog© microplates. The microbial communities immediately downstream from the plant differed from those before the plant and further downstream. The results suggest that effluent from wastewater treatment plants temporarily alters microbial communities but this effect does not persist further downstream.

## NOTES

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## EFFECTS OF PERVIOUS CONCRETE ON POTENTIAL ENVIRONMENTAL IMPACTS FROM ANIMAL PRODUCTION FACILITIES

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Studies have shown that pervious concrete has the potential to increase water quality in urban areas as stormwater or wastewater passes through the concrete matrix. Pervious concrete could provide positive results for pollution reduction when used for animal feeding pads, manure storage pads, or floor systems in animal buildings. Runoff from agricultural activities can have negative impacts on the environment; however, little research has been conducted to determine the performance of pervious concrete for uses in these areas. The objective of this study was to provide more information concerning the use of pervious concrete in agricultural settings.

Laboratory tests were conducted on replicated samples of pervious concrete made from two aggregate sources (river gravel and limestone) with two size fractions of each aggregate. Compost composed of beef cattle manure and bedding was placed on top of the pervious concrete specimens and one liter of water was filtered through the compost and pervious concrete for two separate daily leaching events. T- tests indicated that the mass of compost retained on the surface of the pervious concrete specimens was significantly greater when smaller aggregate sizes (#8 river gravel) were used ( $p = 0.012$ ). Nutrient analyses were conducted on the effluent from the compost and pervious concrete and compared to values from an identical test performed by filtering water through compost on a No. 80 wire mesh screen. These tests indicated that filtering the compost effluent through pervious concrete resulted in significant reductions in total nitrogen, soluble phosphorus, and total phosphorus compared to the wire screen. There were no consistent significant differences between the effects of filtering with pervious concrete or wire mesh screen with respect to other analytes (e.g. dissolved organic carbon, ammonium, nitrate, and nitrite). Effluent BOD levels from the compost and pervious concrete for both daily leaching events (38.7 and 42.5 mg/l, respectively) averaged above typical allowable wastewater concentrations of 30 mg/l. Use of the pervious concrete for filtering resulted in significantly higher pH (9.3) ( $p$  value  $<0.0001$ ) compared to the effluent pH from the wire mesh screen (7.7).

Weekly rainfall simulations were conducted after manure was applied to the surface of pervious concrete specimens. The effluent from the manure and pervious concrete was tested for five-day BOD, dissolved organic carbon, ammonium, nitrate, nitrite, total nitrogen, soluble phosphorus, and total phosphorus. Statistical analysis indicated that significant increases and decreases can occur in these analyte concentrations after rainfall

events. The highest concentrations of some analytes (five-day BOD, nitrate, total nitrogen and total phosphorus) in the effluent occurred after the first rainfall simulation. Maximum concentrations for other analytes (DOC, ammonium, nitrate, and soluble phosphorus) occurred after subsequent rainfall events. Further analysis of the effluent indicated a significant decrease in fecal coliform concentration one week after the initial rainfall simulation. Ammonia and carbon dioxide emissions from the manure and pervious concrete specimens were also monitored for a five day period following three weekly rainfall events. Results indicated that the pervious concrete was capable of providing an environment where ammonia and carbon dioxide could be volatilized. The carbon dioxide emissions indicate microbial activity where immobilization of nutrients and decomposition of the manure could occur. Therefore, additional nutrients could be retained by microorganisms in animal waste deposited on the pervious concrete surface.

## ASSESSMENT OF COAL WASTE IMPACTS ON THE MUNICIPAL WATER SUPPLY IN MARTIN COUNTY, KENTUCKY USING HOT WATER TANKS

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In 2000, a breach in a coal waste impoundment in Martin County, KY released over 300 million gallons of coal sludge and black water into area waterways. Government and corporate sponsored agencies conducted impact assessments. Local residents were dissatisfied by the conclusions reached by the environmental firms under subcontract by the coal company. In 2005, the Kentucky General Assembly authorized money to independently assess the spill's impact on the human and natural environment, especially the local water supply. These studies were conducted with local citizen participation and oversight to ensure the transparency and reliability of data thus generated. Water taken from the drain valve of hot water tanks was used to test for possible accumulation of heavy metals from the municipal water supply. Since sediment and precipitates may accumulate in the tanks from the moment the tank is installed, they may reflect historical accumulation either through long term low level buildup up or though high level spikes that may periodically occur. Fifty-six water heaters were analyzed in Martin county with an additional 30 and 33 samples taken from Pulaski and Madison Counties, respectively, for comparison. Metals analyzed included mercury (Hg), arsenic (As), barium (Ba), cadmium (Cd), chromium (Cr), cobalt (Co), copper (Cu), iron (Fe), lead (Pb), manganese (Mn), and selenium (Se). The type of water heater, type of usage, and usage volume were recorded. Arsenic, Cu, Fe, and Pb exceeded safe drinking water levels in some cases. Further research is underway to assess relationships between this sort of sample and hot water delivered to the tap and thus available to residents. The concentrations of some metals, including As, Ba, Cd, Cr, Co, and Fe were found to correlate with each other consistently. This may allow one of more of these metals to act as an indicator for a broader variety of metals. No clear relationships were detected between age of tank and metal accumulation. A higher percentage of samples from Pulaski County had safe drinking water exceedences than either Martin or Madison counties. Several metals, including As, Cu, Fe, Hg, Mn, and Pb were significantly higher on average (ANOVA-Scheffe's test) than Martin and/or Madison counties. This method may allow the assessment of water quality provided by different municipal water supplies.

## NOTES

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## STUDY OF SOIL EROSION MODELS AT THE WATERSHED SCALE AND DATABASE DEVELOPMENT

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Soil erosion in watersheds is the process by which particles of minerals, nutrient, and organics are removed from the earth's surface. The particles are transported by water and either settled in-stream temporarily or flushed out of the watershed. Watershed erosion models are used to predict soil transported in a watershed based on size and mass flow of water. The models are developed to understand erosion across different size watersheds, physical features, land-use, and GIS ability, all using the type of model and time of the event. This research focuses upon a comprehensive review of over 50 watershed erosion models and construction of an accompanying database for model selection based on modeling characteristics. The review studies models from small and large scale watersheds. Also, I am looking to link GIS programs to models to assist and understand the topography with erosion. We also would like to learn the erosion of soil when land is being used, such as for mining and agriculture sources. These models are categorized as either continuous or event-based. They are also separated as empirical, semi-empirical, and physically-based models. The significance of this research is that with our database, models may be easily chosen for study sites based on different scales, GIS, land-use, and vegetation. It is anticipated that in the future our database will be used for watershed erosion model selection for solving watershed sedimentation problems.





## EXPERIMENTAL STUDY OF A CORRELATION BETWEEN VARIOUS SOIL PARAMETERS AND RESULTANT EROSIONAL COHESIVE STRENGTH

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Erosion of cohesive sediment (silt, clay, and fine organics) heavily influences surface water quality. Many harmful particles such as organic contaminants and metals are entrained in the sediment/pore-water strata of a given sediment bed. The contaminants adsorb to cohesives due to cohesives' distinct attractive nature. Upon sediment resuspension via erosion, the harmful particles are released into surface water flow and pose a potentially dangerous threat. This danger is especially applicable to Kentucky, as siltation due in large part to cohesive sediment erosion is the leading surface water impairment in the state.

Erosional cohesive strength refers to the magnitude of microscopic, electrochemical bonding present in cohesive sediment. In practical application, it describes sediment bed particles' resistance to resuspension in the water column. Because of the microscopic nature of erosional cohesive strength, it has historically been much harder to test and observe experimentally than its macroscopic counterpart we are more familiar with – mechanical cohesive strength.

The relationship between specific soil properties and resultant mechanical cohesive strength is well defined. Less is known about a similar relationship for fluvial erosional strength which, if developed, could aid in current efforts to mitigate Kentucky's surface water quality concerns. The objectives of this study are to examine this relationship and its controlling properties through a literature review, to develop an equation relating the erosional cohesive strength of a soil to those properties, specifically for sediments typical

of Kentucky, and to test the equation in the lab by engineering soils with desired properties and observing their resultant erosional cohesive strength in an experimental flume. This poster presentation describes our ongoing methods and preliminary results to complete the described tasks.

## STUDY OF SEDIMENT AGGREGATES IN THE KENTUCKY RIVER BASIN

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Fine sediment in streams impacts several aspects of water quality including (1) transport of pollutants such as heavy metals, (2) directly and indirectly causing fish kills, and (3) disrupting intake systems of water treatment plants. It is well known that sediment largely travels as composite particles called aggregates or flocs. Despite their significance, a conclusive theory of the factors and processes impacting sediment aggregates in streams has not been substantiated. Further, modeling tools that predict the size of sediment aggregates within the watershed setting do not exist. The objective of this project was to sample sediment aggregates from streams in the Kentucky River Basin and study their size and organic matter characteristics in order to better understand the factors and processes impacting the aggregates. Size is an important characteristic of aggregates because it is directly related to whether the particle will settle or remain suspended for subsequent transport downstream. Organic matter, such as algae, is an important characteristic because it directly impacts aggregate size. Microbial organisms excrete exopolymeric substances (EPS) which have been described as the most important characteristic affecting aggregate size. In addition, organic matter was studied because it is related to a number of environmental issues (e.g., fish habitat, carbon cycling). GIS methods were also employed to determine watershed characteristics such as slope, land use, climate, and soil properties that could potentially affect the aggregates. Three regions within the Kentucky River Basin were chosen to represent different types of land use, because it is hypothesized that land use will impact the processes controlling aggregate size. The South Elkhorn watershed in Lexington, Kentucky was chosen because of its urban and agricultural regions and was broken up into agricultural, urban, and mixed sub-watersheds. The Red River watershed was chosen because of its forested and agricultural land uses. In addition, the Letcher County region was chosen because of the mining and forested land uses and the watershed was broken up into sub-watersheds

with land uses including active mining, recently reclaimed, reclaimed more than 10 years ago, and old growth forest. To study the aggregates, samples are collected in sediment traps and brought back to the lab where a sub-sample is taken for aggregate analysis with an inverted microscope. Algal counts are made, and image analysis software is then used to analyze the size distribution of the aggregates in each sample. Results from the GIS analysis reveal that land use is the most varied characteristic in the sub-watersheds, with other characteristics remaining fairly constant between sites. Algal counts will show trends which may correlate with the trends observed through the aggregate size analysis. Spatial and temporal trends can also be identified with the algal counts and aggregate size analysis. These results will provide an important step towards developing a model to predict sediment aggregate size at the watershed scale.

2001 vs. 1992 LAND COVER CHANGE PATTERNS OVER A FIXED SPATIAL INTERVAL USING THE USGS KENTUCKY CLIMATE DATA GENERATOR OF THE KENTUCKY WATERSHED MODELING INFORMATION PORTAL (KWMIP)

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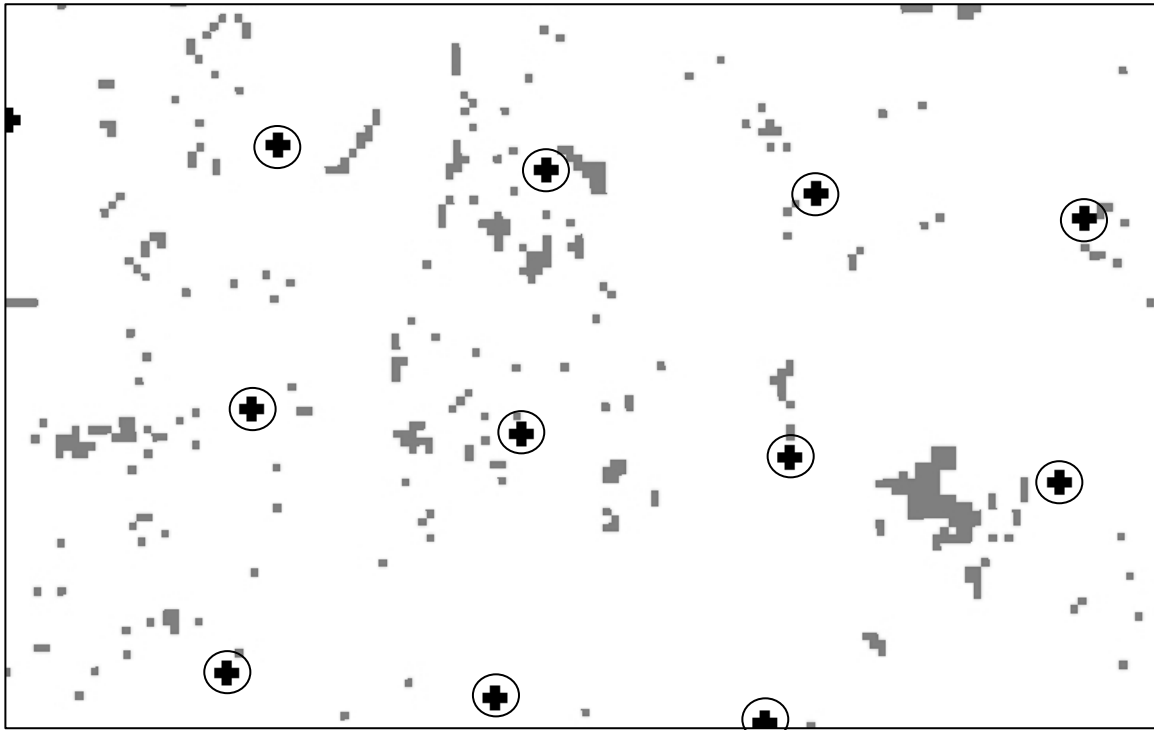
In September, 2004, The Kentucky Commonwealth Office of Technology (COT) was awarded \$750,000 from the US Environmental Protection Agency (USEPA)'s Environmental Information Exchange Network (EIEN) to develop the Kentucky Watershed Modeling Information Portal (KWMIP). This 2-year project will develop a web-based portal to quickly and accurately deliver current, and appropriately formatted, watershed model input data for selected models.

The US Geological Service (USGS) has been continuing work on development of the KY Climate Model (KCM) for temperature and precipitation. The model applies and enhances a spatial regression approach that has been applied in New Jersey and Colorado. The spatial regression model equations have been established and work on the user interfaces is ongoing. The KCM will be accessed through KWMIP to provide daily temperature and precipitation model results in model-ready format. Climactic data are interpolated for nodes along a 1-km grid.

The 2001 vs. 1992 Anderson Level I land cover change mask was produced by USGS (Coan et al., 2006) and processed to eliminate image clutter. The KCM 1-km grid was used to characterize spatial variability of change at pre-determined intervals across several watersheds of interest (Figure 1).

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*Figure 1.* The Kentucky Climate Model 1-km grid (indicated by encircled crosses) overlaying areas of land cover change between 1992 and 2001. Approximate scale: 1:30,000; North direction is up.

## MONITORING SOIL MOISTURE FOR EFFICIENT USE OF IRRIGATED WATER ON SELECTED GRASS LAWNS

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Soil moisture content is one of the factors that controls the infiltration capacity of soils. Precipitation and irrigation increase soil moisture which in turn reduces infiltration capacity. This results in increased runoff during subsequent storm events. Increased stormwater runoff may cause adverse environmental problems such as increased soil erosion, increased bed and suspended loads in streams, and increased non-point source pollution. Monitoring soil moisture on irrigated plots can be used as a guide for efficient use of irrigated water. Thus, irrigation systems only will be turned on when soil moisture falls below a threshold value for the respective soil type. However, landscapers at Northern Kentucky University (NKU) schedule the irrigation of grass lawns without taking into consideration the level of soil moisture. This has resulted in irrigation of the lawns during or immediately after a heavy storm event.

Effective monitoring of the soil moisture of irrigated fields has been shown to help in controlling cost of irrigation and conserving valuable resources. This can be achieved by using instruments such as tensiometers and neutron probes to monitor soil moisture (Manning, 1992). On an irrigated field such as a grass lawn, the ideal condition will be to maintain soil moisture between field capacity and wilting point. The objective of this study is to investigate the effect of soil texture and slope on the amount of irrigated water used on selected grass lawns on NKU campus at Highland Heights, Kentucky. The grass lawns were selected based on low slope (0 to 10<sup>0</sup>), medium slope (10<sup>0</sup> to 15<sup>0</sup>), and high slope (more than 15<sup>0</sup>). Two plots were selected for each slope category. The soil texture of each grass lawn was determined by performing standard particle size distribution analysis of samples taken during the installation of the tensiometers. A survey instrument and GIS software were used to analyze the slopes. The tensiometers were monitored daily and NKU Grounds Department was advised to irrigate those plots only when the soil moisture fell below a specified threshold level. The threshold value was between 70 and 80 centibars for the range of soil textures at the site. Temperature and precipitation data were gathered from NKU's Department of Physics and Geology weather center and the Northern Kentucky Airport Weather Station.

The second low slope plot (LSII) has the highest percent sand of 40% whilst the second medium slope (MSII) has the lowest sand of 30%. Although the textures of the low slope plots are significantly different, there was not much difference between the moisture readings. However, a slight difference in the texture of the high slope plots tends to affect water infiltration and moisture retention capacities. It takes longer for water to infiltrate the finer grained, high slope plot but it retains the moisture longer once it is saturated. Air temperatures of 85<sup>0</sup> F and above were the controlling factor as all plots dried faster after irrigation or precipitation. Overall, the soil moisture monitoring resulted in less irrigated water use; less than half the normal amount.





## NITRITE REDUCTION BY FE(II) ASSOCIATED WITH KAOLINITE

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The microbial reduction of solid Fe(III) hydr(oxide) minerals by iron reducing bacteria is an important process in biogeochemical cycling of nutrients and trace metal contaminants. The presence of  $\text{NO}_3^-$  can inhibit the net Fe(III) reduction to Fe(II) under anoxic conditions. There are several mechanisms proposed to explain such phenomena (Matocha and Coyne, 2007). One proposed mechanism involves simultaneous  $\text{NO}_3^-$  and Fe(III) reduction coupled to chemical reoxidation of Fe(II) to Fe(III) by  $\text{NO}_2^-$ , the intermediate of  $\text{NO}_3^-$  reduction (Komatsu et al., 1978; Obuekwe et al., 1981).

A majority of the Fe(II) produced during microbial Fe(III) reduction exists in precipitated or sorbed forms. For example, Kukkadapu et al. (2001) reported that in Fe(III) oxide-rich subsoils with mixed mineralogy, dissolved biogenic Fe(II) adsorbed strongly to kaolinite. Accordingly, the objective of this study was to investigate the role of Fe(II) associated with kaolinite in the reduction of  $\text{NO}_2^-$ .

Nitrite was added to stirred kaolinite suspensions under anoxic conditions to simulate field conditions where Fe(III)-reducing conditions occur. It was found that nitrite was reduced rapidly by Fe(II) associated with kaolinite when compared with solutions devoid of kaolinite. One of the major products of nitrite reduction was nitrous oxide ( $\text{N}_2\text{O}$ ), an important greenhouse gas. In the process, Fe(II) was reoxidized to Fe(III). This supports the chemical reoxidation pathway of Fe(II) by  $\text{NO}_2^-$  in contributing to the inhibition of Fe(III) reduction. This research is a timely pursuit given the high costs of N fertilizer and the desire to protect water resources from elevated  $\text{NO}_3^-$  levels.

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## EUTROPHIC CONDITIONS IN THREE NORTHERN KENTUCKY STREAMS

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Census Bureau data indicates that the Northern Kentucky region, with a population growth of more than 13.5% since 2000, is rapidly transforming from an agricultural to an urban landscape. With urbanization comes pollution and water quality degradation. Eutrophication, a highly productive condition caused by excess nutrient loading from agricultural runoff and/or treated sewage effluent from urbanized areas, causes noxious algal blooms promoting diurnal dissolved oxygen and pH swings that degrade water quality and aquatic habitat integrity. Water quality declines when impervious surfaces reach 10% of watershed area and at 25% channels incise, banks erode and sediment load is transported downstream where it settles out and destroys physical habitat (Pelley, 2004). Ultimately, though agriculture does contribute to nutrient enrichment, urbanization is currently the biggest threat to local and regional water resources. Urbanization leads to impaired chemical and physical conditions that negatively impact the biological component of surface waters. Though degraded water quality associated with both agricultural and urban land use is well documented, no data exist that describe the trophic state of area streams or land use impacts.

The United States Environmental Protection Agency (USEPA) developed nutrient and eutrophic response variable criteria to prevent nuisance algal growth (USEPA 2000a, 2000b). These criteria are specific to streams with similar geology, topography, land use, and nutrient concentrations (*i.e.* subcoregion), and serve as a starting point for states to delineate their own criteria. For each subcoregion, recommended limits were published for N, P, sestonic and benthic algal biomass (*i.e.* primary eutrophic response variables), DO, and phosphorus storage in benthic algae (*i.e.* secondary response variables) (USEPA, 2000a; 2000b).

Given the current population growth and expansion of urbanized areas in northern Kentucky, it is imperative that we understand as much as possible about the impact of urbanization on local streams. To that end, we are assessing nutrient concentrations and eutrophic response variables in three Northern Kentucky streams. The year-long study, still in-progress, began in April 2006 and will continue through March 2007. There are three main objectives: 1) to document the trophic status of 12 Mile, Doe Run and Banklick Creeks by sampling bi-weekly and comparing data to U.S. EPA criteria for water quality protection; 2) to assess the relationship between trophic indicators: pH, dissolved oxygen, conductivity, nitrogen and phosphorus concentrations, sestonic and benthic algal biomass, and phosphorus concentration in benthic algae in the sampled streams; and 3) to examine the potential for developing a model of water quality parameters that quantifies land use impacts on northern Kentucky streams.

Results presented here indicate that nutrient concentrations, nitrogen and phosphorus, exceed U.S. EPA standards > 70% of the time (one tailed t-test comparison of data to U.S. EPA recommended values,  $p = 0.05$ ). As would be expected with eutrophic waters (*i.e.* nutrient enriched), sestonic and benthic algal biomass, a primary eutrophic response, exceeded U.S. EPA criteria of 5.37 ug/L chlorophyll a and 150 mg chlorophyll a per m<sup>2</sup>, respectively. Periphyton phosphorus storage, a secondary response to eutrophication, also exceeded U.S. EPA criteria of 20 mg/m<sup>2</sup> nearly 85% of the time ( $p = 0.05$ ). We conclude that all three Northern Kentucky streams are eutrophic and that the biological responses to eutrophication detected thus far are a signature of land use impacts on water quality.

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## THE BIG DIP: GEOGRAPHIC DISTRIBUTION OF HIGH METALS OBSERVED IN EASTERN KENTUCKY HEADWATER STREAMS

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The "Big Dip" was a diagnostic sampling of 917 headwaters streams in southeastern Kentucky conducted in the summer of 2006 by 30 volunteers and 6 paid staff in a collaborative project between the Eastern Kentucky Environmental Research Institute, and Head of Three Rivers Project, which staffs an AmeriCorps\*VISTA member in Letcher County as part of the U.S. Office of Surface Mining's Appalachian Coal Country Watershed Team.

The interdisciplinary community-based research project integrated geography and chemistry to establish baseline data on a suite of analytes that could be tested at relatively low cost and using simple field collection methods (e.g., multi-parameter probes and test strips) over a wide geographical area. The field team endeavored to take a water sample at every reasonably accessible first-order stream in a six-county area of eastern Kentucky (Breathitt, Harlan, Knott, Leslie, Letcher, and Perry counties). The samples are concentrated primarily in the Kentucky River headwaters, and Letcher County portions of the Cumberland River (Poor Fork) and the Big Sandy River (Levisa Fork.)

Between June and September, 917 sites were sampled for pH, conductivity, temperature, alkalinity, hardness, nitrite, nitrate, and iron and mapped using GPS units. Among the nine parameters examined, three stood out as potentially good indicators of threats to stream health—conductivity, iron, and pH. While about a third of the samples were in the good or normal range of these parameters, two out of every three sites registered an extreme value of at least one—and often more than one—of these three basic water quality indicators (conductivity > 500µhos; iron > 1 ppm; pH > 8.2 or <5.0).

In addition, a full metals analysis of 36 metals and metalloids was conducted for 21 samples using the ICP at the Environmental Research Training Laboratory (ERTL) at the University of Kentucky. High levels of aluminum, manganese, and iron were identified in many of these samples. These high metals are consistent with acid mine drainage that originates from abandoned mine lands—or what EPA considers “preexisting discharges.”

While several simultaneous analyses of these data are ongoing, this presentation will focus on the geographic distribution of these samples where high metals were observed. Using a GIS-based cluster analysis approach, the geographic distribution of these high metals observations is examined to determine whether or not they are associated with abandoned mines. Furthermore, the “analytic signatures” of the basic parameters that were collected at all 917 sites is compared with these 21 metals analysis sites to determine whether a pattern can be discerned that might be used to identify additional sites that should be tested for heavy metals.

## ASSESSMENT OF AGRICULTURAL BEST MANAGEMENT PRACTICES IN THE BRUSHY CREEK WATERSHED

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The Brushy Creek watershed is located in the south central Kentucky counties of Lincoln, Pulaski, and Rockcastle. This watershed encompasses approximately 29,000 acres located in the Mississippian Plateau physiographic region of Kentucky. This boundary includes the four 14-digit subwatersheds of Upper Brushy Creek, Lower Brushy Creek, Bee Lick Creek, and Clifty Creek. Brushy Creek is a major tributary of Buck Creek. Buck Creek and its tributaries have been a primary focus for the Kentucky Chapter of the Nature Conservancy as they are home to more than 30 species of freshwater mussels (nine of which are endangered or of state concern), 77 species of fish, and one endangered bat species. The threats to this outstanding resource water are incompatible forestry practices, livestock production practices, crop production practices, invasive species, recreational vehicles, and unimproved creek crossings (The Nature Conservancy, 2006).

Buck Creek and its tributaries have been an area of interest for federal and government programs that provide funding for agricultural best management practices (BMP). Buck Creek was a designated United States Department of Agriculture (USDA) Environmental Quality Incentives Program (EQIP) priority area in 2001 and 2002. There has also been funding from the US Fish and Wildlife Service Partnership for Wildlife Program, Kentucky Division of Conservation's State Cost Share Program, and federal Farm Bill programs such as the Continuous Conservation Reserve Program (CRP).

Agricultural best management practices (BMPs) can be defined as methods or a system of methods that are implemented for the reduction of non point source pollution in agricultural watersheds. There is much documentation to attest that these methods are of great value at the small watershed (field) scale, but there is little existing research of effectiveness at the medium or large watershed scale. Spatial scale has a great influence on the modeling of non point source pollution and BMPs. The small (field) scale approach has been successful in modeling the upstream-downstream effects of BMPs on small plots of land such as large farms or groups of small farms. The effects of BMPs at the medium watershed scale, such as that of a small tributary stream or at the large watershed scale, such as a 14-digit HUC watershed is still largely unknown. This research project used water quality and statistical analysis along with geotechniques to assess the effects of BMPs in the large-scale (29,000-acre) Brushy Creek watershed.



Water samples have been collected in the Brushy Creek watershed since May of 2006. These samples have been collected from 25 surface water and 5 groundwater sites. The samples have been analyzed for nutrients (nitrogen, nitrogen ammonia, total phosphorus, and orthophosphate), total coliforms, and escherichia coli. Dissolved oxygen, water temp, pH, and specific conductance measurements were collected in the field using a YSI 556 multi-probe instrument.

Results of the preliminary statistical and geostatistical analysis relating water quality to BMP implementation at the watershed scale will be discussed. While data collection is still in process, preliminary analysis indicates that, at the watershed scale, BMPs may not have a significant influence on water quality. Furthermore, the karstic nature of the watershed and the resulting ground-surface water interactions may reduce the effectiveness of some BMPs—particularly those that aim to restrict cattle from creek bank access by providing alternate watering supplies if the practice leads to cattle congregating in very karstic areas where pollutants are likely to be transported quickly to the stream network via groundwater.

## GROUND-TRUTHING REMOTELY SENSED DATA IN A SMALL WATERSHED ON THE URBAN/RURAL FRINGE

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Accurate land use data is essential to the study of how land use practices affect the quality of receiving waterways, but obtaining very high accuracy at the large scale—the scale necessary to link land use with water quality in a small watershed—can be prohibitively expensive. The purpose of this study was to test a method of generating a land coverage at reasonable cost by combining the NLCD dataset, the National Agriculture Imagery Program (NAIP) digital orthophotography, and a limited number of field observations to produce a revised polygonal land use layer. The study site is the drainage area for Wilgreen Lake—a small manmade lake on the outskirts of Richmond, Kentucky.

At present, a common and inexpensive land cover layer is the National Land Cover Dataset (NLCD), a raster-based coverage derived from 1992 Landsat thematic mapper imagery at a 30-meter resolution. While it is good for the state scale, the 30-meter resolution and rasterized pixellation is much less accurate at the small watershed scale where many land management decisions take place.

During fall 2006, a geographic information system (GIS) was used to overlay the NLCD layer with the NAIP imagery, highlighting conflicts in landscape type primarily by shape. Using the maps generated, a series of field sampling sites were identified, and observations of landscape type were made at these locations and hand-drawn over the printed map. A revised land coverage was then generated in polygonal form using the same 21 land cover categories from the NLCD.

While the methodology was generally successful at producing a high-resolution land coverage at relatively low cost, several problems were encountered in the process that should be noted. First, pixels were not always aligned with the NAIP land cover images. Second, when the newly generated coverage was compared to the NLCD, some forested areas appeared misplaced due to pixilation imprecision. Visual inspections of the watershed also identified several misclassifications in the NLCD imagery—notably that “pasture” was incorrectly classified as “developed open space”—a classification error that could have major impacts on a watershed study. Finally, changes in land coverage have occurred since the imagery was taken in 2001. These last two issues highlight the importance of supplementing the GIS overlay process with adequate in-field ground-truthing to derive a sufficiently accurate land coverage for use at the watershed scale.

## NOTES

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