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DETERMINATIONS OF BIOAVAILABLE FRACTIONS IN THE ASSESSMENT OF METALS IN BIG AND LITTLE BAYOU CREEKS DUE TO EFFLUENT DISCHARGES ORIGINATING FROM THE PADUCAH GASEOUS DIFFUSION PLANT, MCCRACKEN COUNTY, KENTUCKY

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The hypothesis of this study was that even though metals may be present in the water column, not all of the metal concentration is available to aquatic organisms; therefore risk assessment based solely on total recoverable metals would overestimate environmental impacts. During 2000 through 2005, chemical characterizations and biomonitoring were conducted on Big and Little Bayou creeks, which were impacted by effluents from the Paducah Gaseous Diffusion Plant, McCracken County. Water, sediments, and sentinel fish samples were analyzed for nine metals (Ag, Be, Cd, total Cr, Cu, Fe, Ni, Pb, and Zn). Metal body burdens in stoneroller minnows (*Campostoma anomalum*) were used to develop multipliers to convert total recoverable water metal concentrations to bioavailable metal concentrations. Detections of Be, Cr, and Pb in waters from Big Bayou creek tended to be sporadic, while Ag, Cu, Ni, and Zn were consistently detected, indicating chronic contamination by these metals. Determinations of bioavailable fractions for stations in the effluent receiving zone (ERZ, stations BB4-BB7) indicated that for Cd, Cr, Fe, and Pb, less than half of the water column metal was bioavailable to stoneroller minnows. In contrast, 60 % of Ag, 73 % of Cu, and 64 % of Zn were bioavailable to stoneroller minnows in the ERZ. In Big Bayou creek, stoneroller minnow Cu body burden correlated with water column Cu, but poorly with sediment Cu, indicating that most of the copper uptake was from the water column. Unlike results from Big Bayou creek where only 3 metals were >59 % bioavailable, most metals found in Little Bayou creek water were >70 % bioavailable. In general, most of the sediment metals from both streams did not correlate with stoneroller minnow body burden, possibly due to high binding of the metals to the sediments. No seasonal changes were observed in metal concentrations. Results from this study support the hypothesis that total recoverable metal concentrations overestimated biologically available metal concentrations. Several developing models, such as the Windermere humic aqueous model (WHAM) and the biotic ligand model (BLM) being used for predicting bioavailable metals are limited by their focus of a single route of exposure, namely respiratory exposure. These models do not take into account other important routes, such as dietary exposure. The use of stoneroller minnows as *in-situ* sentinel monitors proved to be a valuable concept and allowed the study of metal behavior in stream systems under real-world conditions.

NOTES

CREATION OF A CATALOG OF ENVIRONMENTAL MERCURY DATABASES IN KENTUCKY

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Mercury is a potent toxin to humans, with the nervous system of the developing fetus being the most susceptible to its effects. A primary route of exposure is ingestion of mercury through fish consumption. In Kentucky, mercury levels in fish tissue are consistently out of compliance with the Clean Water Act, resulting in fish consumption advisories. A better understanding of the relationship between environmental mercury sources and mercury levels in fish is necessary to plan strategies to reduce these levels in order to protect human health.

This project is the first step in evaluating the problem of mercury levels in fish. A number of agencies collect information on mercury levels in the environment. However, there is no coordination between agencies for evaluating these data or a common platform for assessing the data. Databases were identified by contacting federal, regional, state, and local agencies, and a catalog was created. For each database, the catalog includes a review of the sampling strategy, collection method, form(s) of mercury, collection sites, date range, frequency of sampling, and data format. Quality assurance and quality control methods were reviewed to evaluate the reliability of the data and these processes are included in the catalog. The entries include who has access to the database, and how the database is accessed. The catalog also contains maps of mercury emission sources and sampling sites.

Information on airborne mercury is collected and maintained by three agencies or programs: the Toxics Release Inventory (TRI) is a regulatory database that includes estimates of mercury emissions from coal-fired power plants; the Kentucky Division for Air Quality (DAQ) samples for mercury by two different methods, wet deposition and continuous ambient air monitoring; and the Mercury Deposition Network (MDN) is a national collaborative effort that monitors wet deposition of mercury across the nation.

Mercury monitoring in water is done in several different media: the water column-both groundwater and surface waters, fish tissue, and sediment. In addition, the water column can be monitored for total mercury and dissolved mercury. Several agencies monitor for mercury in water in Kentucky. The Kentucky Division of Water (KDOW) is responsible for water quality in the state, and has the most comprehensive monitoring program. KDOW monitors for total mercury in the water column, fish tissue, and sediment. The Ohio River Valley Sanitation Commission (ORSANCO) is an interstate agency that monitors the Ohio River and its tributaries. ORSANCO monitors the water column for both dissolved and total mercury. Fish tissue is monitored for methylmercury. The Metropolitan Sewer District (MSD) is responsible for the monitoring of surface waters in Jefferson County, Kentucky. MSD monitors for total mercury. Lexington Fayette Urban County Government (LFUCG) had limited water column, sediment, and fish tissue sampling from 1992 to 2003. The Kentucky Division of Waste Management (DWM) does some groundwater monitoring around landfills to determine if mercury has leached out of these sites. The Kentucky Geological Survey (KGS) has a more comprehensive groundwater monitoring program and maintains a searchable database on the internet that reports groundwater mercury data from a number of agencies.

This study found that data collected for regulatory purposes are not always collected in a manner that is practical for research or maintained in a platform that is readily accessible. In addition, a lack of uniformity across agencies in the species of mercury sampled, variation in quality control, and differences in ease of access were findings that make using these data for research more difficult. Quality control and quality assurance methods were reliable for state and regional agencies, but were inconsistent in local agencies. However, state and regional agencies often monitored for different species of mercury, resulting in data that are not comparable. For several databases, geographic information was kept in separate files, complicating the task of performing geographic analyses. The Kentucky Department of Environmental Protection (DEP) is transitioning to a new database platform, COMPASS, that will facilitate the access of data kept by all divisions within the department. This platform will improve the ability to analyze information between the various media and provide geographic information of sample sites instantly.

At this time there is no coordination of sampling strategies between the DEP divisions. The DAQ monitors for mercury based on emission sites, while also monitoring one background site. The KDOW monitors major rivers and waterways monthly, and samples each of its five major watershed regions more intensely on a five year rotating basis. Coordination of sampling strategies between the various media is necessary to perform meaningful analyses to evaluate the fate and transport of mercury in the environment. The DEP has begun to bring the DAQ, KDOW, and DWM together to discuss the issue of mercury and how to address this problem. This may result in a shared strategy for monitoring mercury which is a necessary first step in devising a strategy to reduce its impact in the environment.

Significant amounts of mercury sampling data are available from federal, regional, state, and local agencies which monitor for regulatory purposes. With attention to quality control and quality assurance, coordinated sampling strategies, and attention to the manner in which these data are maintained and stored, these data could be used to help devise strategies to reduce levels of mercury in fish. The DEP has begun to work toward this objective by beginning to transition to COMPASS as a database platform, and bringing the various divisions together to discuss a coordinated strategy for addressing the problem of mercury levels in fish.

USING NITROGEN ISOTOPES TO TRACE NUTRIENT INPUTS TO A EUTROPHIC LAKE, WILGREEN LAKE, MADISON COUNTY, KENTUCKY

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Wilgreen Lake is a dammed lake that has been classified as nutrient-impaired (303d list) by the EPA and State of Kentucky. The lake is moderately-sized covering 169 acres (0.7 km^2), and drains a watershed with residential developments, cattle pasture, modified woodlands, and some industrial/urban usage in the city of Richmond. The principal tributaries are Taylor Fork and Old Town Branch that meet to form the trunk of the lake approximately one mile in length. The upper reaches of Taylor Fork are adjacent to a densely-packed (quarter-acre lots) housing development with septic systems. Old Town Branch drains cattle pasture and residential areas. Residences within these developments, while also served by septic systems, are more sparsely distributed than residences within developments adjacent to those of Taylor Fork. An ancillary tributary flowing into Pond Cove is intermittent, and drains cattle pasture and one small housing development. Recognizing and quantifying potential nutrient sources is critical to any remediation efforts in decreasing nutrient input to the lake. We hypothesize that significant nutrient input occurs from the septic systems adjacent to the shallow lake waters of Taylor Fork. We use stable nitrogen isotopes (^{14}N and ^{15}N) as a tracer in characterizing organic sources of nitrogen entering lake waters, and in characterizing organic sinks of nitrogen residing in the lake system.

We measure the carbon-to-nitrogen ratio, carbon isotopic composition ($\delta^{13}C$), and nitrogen isotopic composition ($\delta^{15}N$) of organic matter held within potential nutrient sources and sinks within the Wilgreen Lake system. Potential sources include fertilizers, bovine fecal matter, human effluent from septic systems, and “natural” organic material. Sinks include plankton, macroalgae, macrophyta, and organic matter within sediments.

Our samples are being measured at press time. The fundamental assumption of the test of our hypothesis is that $\delta^{15}N$ values of nitrogen sinks should reflect that of their source. With knowledge of the nitrogen isotopic composition of nitrogen sources, we may be able to recognize gradients within the nitrogen sinks of the system. Consequently, our samples of plankton, macroalgae, macrophyta, and sedimentary organic matter are taken over the entire expanse of lake.

NOTES

USING MICROBIAL DISTRIBUTION AND ABUNDANCE IN A EUTROPHIC LAKE AS A TRACER FOR NUTRIENT INPUTS, WILGREEN LAKE, MADISON COUNTY, KENTUCKY

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Wilgreen Lake is a eutrophic lake that has been listed on the EPA's 303d list as nutrient impaired. Potential sources of this impairment are likely from humans, cattle manure, and fertilizers. We suspect that the majority of nutrients originate from human sources, namely from septic tank effluent emanating from key housing developments ringing the lakeshore. We test our hypothesis using two independent methods: a combination of nitrogen isotopic analyses; and conventional microbial assays (*Escherichia coli*) and RT-PCR techniques (*Bacteroides*). This paper examines only our microbe data, principally during the 2007 field season.

For the microbial tracer study, we took water samples at 19 sampling locations on 4 occasions for both tests. Sampling occurred within three tributaries of the lake (Taylor Fork, Old Town Branch, and Pond Cove), as well as its trunk. Sampling spanned about 2 months from 26 June to 15 August with the last 3 sampling events occurring at roughly two-week intervals. We measured the abundance of *Escherichia coli* using IDEXX methods, which approximates the number of colony-forming units (CFU) per 100 mL in terms of most probable number (MPN). Corresponding sub-samples slated for potential PCR analysis were stored at -40^{o}C until all microbial assays were completed. Then we chose PCR candidates on the basis of elevated *E. coli* levels, and the probability of differing source contributions.

E. coli levels are generally elevated (up to >2419 MPN) at sites adjacent to septic tank clusters at the upper reaches of Taylor Fork. The second main tributary, Old Town Branch produced *E. coli* levels consistently lower (3 - 410 MPN), and *E. coli* levels are also low (1 - 24 MPN) in the third lake tributary that dominantly drains cattle pasture. There is a decline in microbial abundance (1 - 20 MPN) distal to these populated areas, and as you approach the main trunk of the lake. This suggests that the principal source of microbial input is from septic systems; however, we cannot eliminate the possibility that fecal microbes are introduced into the lake via inflows. For example, a sewage pumping station exists immediately upstream of the septic tank clusters of Taylor Fork. Thus, high microbial abundance often observed here may also be due to this alternative input, which is likely to enter the lake system during periods of significant rainfall.

We used quantitative PCR analysis to measure *Bacteroides* abundance, and to distinguish between human and cattle sources of this microbe. We measured 14 samples with most samples (11) taken on 1 August 2007, and ancillary samples (3) taken on 15 August. Total fecal microbe concentrations in all samples targeting all *Bacteroides* species ranged from 45 mg/L to 142 mg/L. Unlike other studies, there was no apparent relationship between the concentration of all *Bacteroides* species and that of *E. coli*. The reasons for this result are unclear.

We also attempted to quantitatively assay the proportion of *Bacteroides* contributions from specific sources, namely human and bovine fecal matter. Although fecal contamination was measured in all 14 samples, only 1 sample had significant amounts of human fecal contamination (21%) as measured by the human-associated *Bacteroides* assay. None of the samples had significant bovine fecal concentration as measured by the bovine-associated *Bacteroides* assay. These inconclusive results suggest that either there are other unidentified sources of fecal contamination by *Bacteroides* and/or *E. coli*, or that the prevailing drought conditions skewed our results by not capturing fecal transport effects due to lack of surface and/or groundwater flow.