



WATER RESOURCES RESEARCH GRANT PROPOSAL

(1)**TITLE:** Using Neural Networks to Identify and Quantify Significant Sources of Encysted Protozoa in Watersheds

(2)**FOCUS CATEGORIES:** WQL, SW, NPP, MOD

(3)**KEY WORDS:** Animal Wastes, Bacteria, Viruses, *Cryptosporidium*, *Giardia*, Indicators, Land Use, Fecal Contamination, Agriculture, Risk Management, Water Quality, Public Health, Neural Networks

(4)**DURATION:** 8-1-98 TO 7-31-00

(5)**FEDERAL FUNDS REQUESTED:** \$72,960

(6)**MATCHING FUNDS PLEDGED:** \$147,193

(7)**PRINCIPAL INVESTIGATORS:** Kentucky Water Resources Research Institute

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(8)**CONGRESSIONAL DISTRICT:** 6TH KY

(9)**CRITICAL REGIONAL AND STATE WATER PROBLEM ADDRESSED:**

Pathogenic, encysted protozoan parasites (oo/cysts) of *Cryptosporidium parvum* and *Giardia lamblia* are introduced into surface waters from animal fecal material and human sewage. Indeed, it would seem that their presence in our nation's surface waters is quite ubiquitous, and this presence can threaten the health of the public. Encysted protozoa in watersheds, surface-contacted aquifers, lakes, creeks, and rivers are of concern because they: 1) can persist for long periods (months) in water and on land, 2) are difficult to

isolate reliably from environmental samples, 3) have no reliable indicator for their presence or source, 4) can come from multiple, but not readily distinguishable, sources, 5) are small enough to pass through commonly used rapid-rate, granular filter media water treatment plants, 6) are very resistant to disinfection processes, and 7) have been responsible for many outbreaks of gastrointestinal illness caused by contaminated drinking water. Knowing the potential for harm that encysted protozoa have, and the prohibitive expense of equipping every water treatment facility with 100% effective oo/cyst removal processes, the best option for public protection is prevention. For prevention to be effective however, the significant sources of oo/cysts in each watershed need to be defined and managed, but identification of those sources is not as easy as it would seem. Many of the sources of encysted protozoa in a watershed are not of a point-source nature, being spread by animals that freely roam, and are difficult to identify and remediate due to their diffuse and intermittent nature. However, not all animal wastes have equally large impacts upon encysted protozoan concentrations in watersheds. Some animal sources are known to be worse than others. Human sewage is known to be a prominent source for *Giardia* cysts, while cattle and other animal farming activities are known to be significant sources of *Cryptosporidium* oocysts. Therefore, as large, commercial, animal husbandry and farming operations continue to expand and move into the southeast region, and human sewage continues to be piped directly into rivers and streams, the need for research into water quality and management methods and techniques to effectively identify these sources presence, and their resultant impact on oo/cysts loading, is required.

Water treatment facilities and watershed managers need to have easy, reliable methods and systems for data analysis that can identify when human or animal wastes are impacting a watershed. They need to be able to tell when the most dangerous types of fecal sources are present, be able to prove that the sources are significant so that improvements can be justified, and have an understanding as to the extent of public health risk that may be involved during transient runoff events. It is unreasonable to expect that there would be continuous monitoring by human personnel (the only way currently to reliably both identify and locate potentially high-risk, but transitory animal waste sources) of the potential sources within any given watershed. Therefore an indicator system that can be used at the treatment plant intake and within defined sections of the watershed needs to be developed. Because routine pathogen monitoring for encysted protozoa is not yet economical or practicable, and commonly used indicator bacteria cannot reliably distinguish between different types of animal fecal inputs or indicate the presence of encysted protozoa, surrogates are needed to provide identification and warning of pathogen risk. We propose using a combination of routine sampling for bacteria, with analysis for specific biomarkers of human and animal wastes, with new neural network data analysis techniques to identify and distinguish between various sources of animal and human fecal contamination at a distance from the source. It is our hypothesis that while a single bacterial, bacteriophage, or biochemical indicator may not show significant correlation with animal husbandry wastes and their potential oo/cyst presence, a combination of indicators will. A pattern of indicator presence, or fingerprint, for fecal contamination can be developed that will serve to warn water utilities of increased protozoan loading into treatment plants from different types of

sources and provide an identification system to track the pathogen inputs and facilitate implementation of watershed management measures. This collaborative project intends to employ state-of-the-art methodologies to characterize, quantify, and trace sources of animal and human fecal microbial contamination in watersheds and aquifers in the southeastern states.

(10) EXPECTED RESULTS, BENEFITS AND INFORMATION TO BE GAINED FROM THE PROPOSED RESEARCH:

The information obtained from the proposed research will better characterize the potential risks to human health posed by fecal wastes by better characterizing the sources. The research will provide information the occurrence of *Cryptosporidium*, *Giardia* and other indicator microorganisms in runoff from agricultural animal wastes, as well as the extent to which agricultural and domestic sewage wastes are causing the selected surface and ground waters to be contaminated. Because parasite monitoring is not yet practical for routine water quality monitoring, reliable indicators are needed to detect and distinguish between animal and human fecal waste contamination. The information (data and data analyses) from this study will provide a scientific basis to perform a qualitative risk assessment of parasitic pathogen inputs into the identified surface water sources from agricultural and suburban fecal sources on receiving waters used for various beneficial purposes. This research will provide further data on the relationships between encysted parasites (oo/cysts) and other indicator microorganisms and may well provide a system that will become the surrogate for oo/cyst presence. A system such as the one proposed is needed by farmers, managers, scientists, engineers and regulators to tackle the problem of controlling non-point source fecal inputs into drinking water supplies.

Development of a neural network based indicator system would:

1. Identify the most probable source of fecally originated bacteria present in surface water (human, agricultural animals, or urban animals)
2. Relate the pattern of indicators to the presence, and magnitude of, protozoan cysts and oocysts in surface waters

Establishing this type of indicator system would serve to protect the public health by alerting water treatment plant operators and other health officials of increased parasitic oo/cyst concentrations while providing information about probable sources upon which to initiate control measures.