



Researchers examine emerging harmful contaminants in wastewater effluent in eastern Iowa

Study by Greg LeFevre, University of Iowa; Rebecca Klaper, University of Wisconsin-Milwaukee; Dana Kolphin, USGS; Luke Iwanowicz, USGS

## Why this research was needed

Wastewater, or water that has been used for residential purposes including bathing, toilets, dishwashing, and more, undergoes a treatment process and is released back into the public water system. Streams in Iowa are often highly impacted by treated wastewater, also known as effluent, due to the volume released and the presence of chemicals that living organisms can be exposed to within the aquatic environment. For example, chemicals found in pharmaceuticals have been discovered in fish neural tissue. The presence of contaminants in effluent is widespread in US streams.

As a result, further study is needed to understand the presence of chemical compounds and the impacts of sustained exposures on life within the streams. Additionally, it is important to assess the mixtures of pharmaceutical compounds and the possible negative impacts a combination of chemicals may have on living organisms within the aquatic environment.

The researchers seek to help water resource managers to see the connection between presence of chemicals in wastewater effluent and their complex environmental impacts. This includes enhancing environmental cost-benefit analysis and to improve the risk assessment for chemical contaminants in the environment. In this project, researchers communicated directly with local water resource managers in the project to deliver results and discuss impacts.

## How was it done?

Research occurred at Muddy Creek, an effluent dominated stream in Coralville, Iowa. Muddy Creek is heavily influenced by wastewater effluent from the North Liberty, Iowa wastewater treatment facility. Water samples were taken throughout the project to determine chemicals present in the water as well as their presence as a metabolized form. Measurements were taken at different lengths of the stream to measure their presence and how chemicals changed and intermixed over time and space. In the lab, Zebrafish embryos were exposed to the stream water collected, allowed to develop for approximately two weeks, and then evaluated for developmental delays. Fathead Minnows were also placed directly in Muddy Creek in cages to assess exposures directly within the stream for approximately three weeks. Following exposure, the fish gene expression was examined within a lab to assess disruptions of the endocrine system, which regulates hormone levels such as metabolic rate, as well as other biochemical pathways within the fish.

The goal of this research project is to determine the biological effects of effluent chemicals on fish specimens from wastewater as well as to develop a prediction model for the impacts of future exposures.

## What the researchers discovered

Researchers found that pharmaceuticals in effluent dominated streams change over space and time into complex mixtures. This is due to the variety as well as the volume of chemicals found in effluent. During the three-year project, the researchers found 74 pharmaceuticals, as well as pesticides, and their changing concentrations throughout the length of the stream. When assessing the ecological risk within the stream, pesticides drove a greater risk than pharmaceuticals under both measured and modelled conditions in the study. Researchers looked at concentrations measured under low-flow conditions, and then modeled the concentrations for 'all flows' (which would include conditions influenced by rain runoff). Researchers then compared the concentrations to literature published risk concentrations for different chemicals and different organisms. Different organisms will have different toxic effects to different chemicals. Under both the measured (low flow) and modeled (higher storm flow) conditions, pesticides were the chemicals that drove risk to most organisms-especially aquatic invertebrates. The risk, based on literature published ecotoxicology findings, would be detrimental effects to the organism's ability to function biologically.

Having both a lab and in-field experience enabled the researchers to connect the exposures to effects through analyzing the fish hormones, genetic expression, and the presence of antibiotic resistant bacteria. Researchers conducted a suite of different exposures/effects testing. One of these examined the estrogenicity of the water (hormone disruption), which was not found to match the chemical levels (thus implicating unknown compounds). Researchers also measured transcriptomic impacts, which is the study of the expression levels of mRNAs in a given cell population. The transcriptomic impact included musculoskeletal, cardiac, and metabolic processes, which varied by developmental stage and displayed sensitivity to changes in chemical composition and concentration of effluent, indicating a need for stage-specific biomarkers. Although traditional biomarkers of endocrine disruption were not enriched, a high estrogenicity signal at the upstream site was associated with altered development at 6 days post-fertilization, suggesting the presence of unidentified chemical inputs. This work reveals

associations between chemical mixture composition, developmental stage, and exposure bioeffects and underscores the importance of measuring effects beyond the endocrine system when assessing the impact of bioactive chemicals in wastewater effluent. Ongoing work is investigating neurological genomic analysis.

LeFevre and other project leaders directly communicated findings to the wastewater treatment plant staff. An important finding is that the treatment process works very well for the regulated compounds (far exceeding permit requirements), but the plant was not designed to treat some sorts of emerging contaminants. One important implication is to try to help keep pharmaceuticals and pesticides out of wastewater; in some places this has been accomplished through public education campaigns. Examples include household hazardous waste drop off sites and drug take-back programs.

## What's next?

An important finding was that complex exposure mixture in the streams was driven by interactions with streambed sediment. Project leads will follow up on this by partnering with hydrogeologists to characterize the groundwater-surface water interactions for potential impacts of the chemicals to groundwater. The researchers will also investigate the uptake of chemicals into organisms, and as a result, the foodchain. The researchers will continue to work with North Liberty wastewater treatment plant managers to examine pollutant removal technology to address findings from the study.

A critical finding in the study was the level of neonicotinoid pesticides found in the wastewater. This poses concerns to overall water quality, as well as exposure to humans. Wastewater analysis gives researchers and water managers clues as to unanticipated routes to human exposure. In the study, the city has a separate sewer system, and so pesticides in the wastewater indicate usage in the home, as opposed to outdoors, which is a potential public health threat. Further work will be done to communicate this finding to homeowners and real estate managers.

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