Research Brief 363: Using a New Model to Identify Health-Impacting Metal Mixtures

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You are listening to the Superfund Research Program's Research Brief podcast. This month's brief is titled, "Using a New Model to Identify Health-Impacting Metal Mixtures," and will highlight a new statistical model that can help estimate the neurobehavioral effects of metals found in drinking water. This new model identified six metals, both alone and in mixtures, that consistently affected zebrafish behavior when present in water.

Researchers funded by the NIEHS Superfund Research Program, or SRP, developed the linear mixed-effects model – abbreviated as LMM – a framework for statistical analysis, to quickly and effectively estimate the effects of individual metals and metal mixtures on zebrafish larvae behaviors.

Zebrafish are often used as model organisms to study human neurodevelopment because their nervous system is structured similarly to a human's. Scientists can test effects on zebrafish neurodevelopment by exposing the larvae to certain metals and then measuring their swimming distance.

The researchers, led by Dr.Nishad Jayasundara, a project leader from the Duke University SRP Center, wanted to learn which metals in a mixture were the most responsible for behavioral changes in zebrafish. However, current data analysis methods lacked the accuracy needed to do so, especially with a complex fish behavior dataset. The research team set out to develop a new model to overcome those limitations.

To improve the quality of their analyses, Jayasundara, with support from graduate student Kanchana Dilrukshi and Duke SRP Trainee Ilaria Merutka, designed an LMM that accounted for the following:

- Zebrafish-specific variables, such as the distance each zebrafish traveled;
- Water-specific variables, like the type and concentration of metals in the sample; and
- Swimming distance measurements taken from the same larvae over time, accounting for individual differences among larvae.

They tested the LMM using a dataset from a 2021 study, also led by Jayasundara, that used traditional statistical methods to analyze zebrafish larvae swimming distance after the larvae were exposed to metal-contaminated well water from Maine and New Hampshire.

The LMM identified six possible “bad-actor” metals that consistently affected larval motor activity: lead, cadmium, nickel, copper, uranium, and barium. Lead, cadmium, nickel, copper, and barium were linked to decreased larvae swimming distance, while uranium was linked to increased swimming distance. According to the researchers, these findings indicate that exposure to these metals may be linked to developmental delays or hyperactivity, which may be extrapolated to similar disorders in humans.

Additionally, the LMM identified three metal mixtures – barium, copper, and uranium; nickel, lead, and uranium; and barium, lead, and uranium – that affected zebrafish behavior, even at levels where the individual metals are considered safe.

To validate their model’s findings from the drinking water samples, the scientists then exposed zebrafish larvae to the metals individually – excluding uranium, which was too dangerous to test with – and conducted swimming distance tests. They found that exposure to four of the five metals matched the effects the LMM predicted, indicating that the model is relatively accurate. Cadmium was the only metal that did not match the model, which the authors hypothesize may be because not enough cadmium was present in the samples for an accurate prediction.

According to the researchers, while the previous statistical approaches like the one used in the 2021 study are insightful, the new LMM approach is more sensitive and allows scientists to consider fish behavioral changes over time. The new model enables identification of any significant metal mixtures to determine the most hazardous compounds in a complex mixture.

More broadly, the study shows that LMMs can be an accurate method of analyzing complex data and revealing the potential neurobehavioral health impacts of exposures to pollutant mixtures. Defining “bad-actors” in a mixture is becoming increasingly important because of multiple sources of aquatic contamination, the authors say.

If you’d like to learn more about this research, visit the Superfund Research Program website at www.niehs.nih.gov/srp. From there, click on the Research Brief title under the banner, and refer to the additional information listed under the research brief. If you have any questions or comments about this month’s podcast, send an email to srpinfo@niehs.nih.gov.

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