MDAG.com Internet Case Study 84

Oversimplification of Toxicity and Bioavailability in ML-ARD Studies

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1. Introduction

This MDAG Case Study #84 is driven by overly simplistic statements about toxicity and bioavailability as they pertain to metal leaching and acid rock drainage (ML-ARD) on and around minesites. Two of many overly simplistic statements frequently heard in ML-ARD studies are:

1) water is not toxic because it passed the 96-hour LC50 bioassay test, and
2) for geochemical analyses, the partial digestion of rock, tailings, and other mine wastes by two-acid aqua regia provides an indication of bioavailability of contaminants in the mine wastes.

2. Oversimplification: "water is not toxic because it passed the 96-hour LC50 bioassay test"

Google AI explains,

> "A 96-hour LC50 bioassay is an acute toxicity test that determines the Median Lethal Concentration (LC50) of a substance by exposing fish to various concentrations for 96 hours, with the LC50 being the concentration predicted to kill 50% of the test organisms. This standard method assesses the toxicity of a substance to aquatic life and is used to gauge the hazardous potential of waste, compare the toxicity of different substances, and establish baseline susceptibility for monitoring purposes... Purpose of the Bioassay: ... Used to determine if a waste material is toxic and hazardous to aquatic life."

This is a typical and common explanation of the test. But does it really "determine if a waste material is toxic and hazardous to aquatic life"? From scientific and medical perspectives, the easiest way to show that this statement is overly simplified is by listing some forms of toxicity:

Acute toxicity: Measures mortality within 24-96 hours of exposure to a substance.

Chronic toxicity: Measures mortality and sublethal effects, such as reduced growth and reproduction, over a period of at least seven days.

Subchronic toxicity: Determines the toxicity of repeated exposure to a substance over several weeks or months.

Carcinogenicity: Determines whether a substance can cause tumors by observing animals for signs of toxicity and tumors over a large portion of their lifespan.

Developmental toxicity: Determines the potential effects of prenatal exposure to a substance on a developing fetus.

Genotoxicity: Determines whether a substance can cause DNA damage, chromosomal abnormalities, or genetic mutations.

Reproductive toxicity: Determines whether a substance has the potential to cause birth defects.

Neurotoxicity: Determines whether a substance can cause damage to the nervous system.

Systemic toxicology: Determines the general toxic effects of a substance, including the route of exposure (oral, dermal, or inhalation).[1]

How can a single test like the 96-hour LC50 bioassay "determine if a waste material is toxic and hazardous to aquatic life", as Google AI states, for all types of toxicity and all life forms? It cannot. It only evaluates acute toxicity over a short time for a specific life form.

A human can typically survive without food for more than 96 hours. Does that prove humans do not need food to survive? Does a lack of mortality after a 96-hour LC50 bioassay really prove a water sample is non-toxic? Does it prove there are no other forms of toxicity requiring longer times to be expressed and detected? Does it prove there is no acute toxicity after 100 or 150 hours?

From a legal and regulatory perspective, the 96-hour LC50 bioassay also fails to reliably determine toxicity and impact. For example, Canada's Fisheries Act prohibits a person from carrying out work or activity resulting in the harmful alteration, disruption, or destruction of fish habitat. This Act defines "fish habitat" to include any water frequented by fish and other areas they depend on, such as spawning, nursery, rearing, food supply, and migration areas. Therefore, under this Fisheries Act, the harmful alteration of benthic organisms and the lower part of the food chain on which fish depend (their "food supply") is also prohibited *even in the absence of acute toxicity to fish themselves*.

It is fair to say that the statement "water is not toxic because it passed the 96-hour LC50 bioassay test" is an oversimplification and can in many cases be wrong. Nevertheless, this misconception and error persist to the point it is even repeated by Google AI.

3. Oversimplification: "the partial digestion of rock, tailings, and other mine wastes by two-acid aqua regia provides an indication of bioavailability"

ML-ARD experts have stated, including in public hearings, that aqua regia is a partial digestion and analysis of part of the total amount of each element and contaminant in mine wastes, indicative of

[1] Many thanks to Ms. Jillian Chown for this concise list of some forms of toxicity.

the bioavailability of each contaminant.

To follow a tangent for a moment, these same experts compare their partial aqua-regia concentrations with the near-total concentrations of crustal abundances, and often observe their partial levels are relatively low compared with crustal abundances. They then incorrectly conclude that their low contaminant levels are safer compared to the earth's crust, when in reality they simply dissolved and measured only part of the contaminants in their samples.

The comparison of "oranges to apples" in this case, that is, their partially-dissolved samples to total crustal levels, is not a fair comparison and automatically underestimates contaminant levels in their samples. This is one of many reasons that ML-ARD potential is often underestimated and that predictive errors continue to occur. As stated by many authors for many decades, Nario et al. (2025) reiterated, "the experimental conditions play a relevant role in obtaining reliable information on the mobility and availability of metals".

Returning to a single aqua-regia-digested concentration of an element in a sample as indicative of bioavailability, two simple examples show this is so oversimplified as to be grossly in error. First, what is bioavailable to benthic organisms or terrestrial vegetation can be very different from what is bioavailable to animals or fish. Thus, a single value of an element from an aqua-regia-digested sample is virtually meaningless for bioavailability.

Second, even for a single life form, bioavailability is not a single value of an element due to various exposure pathways. For example, bioavailability in the human digestive system, over approximately a one-day residence time, will be much lower than other exposure pathways. For example, inhaled fine-grained mine wastes with months of residence time within human lungs are subjected to aggressive enzymes and compounds continually dissolving the mine waste so that lungs do not fill entirely during a lifetime[2].

To expand on this issue and to examine it in more detail, Semple et al. (2004 and 2007) stated:

> "It is well known that the portion of a chemical that is either bioavailable or bioaccessible in a given soil or sediment environment can differ substantially between organisms."
>
> "The term bioavailability has been and is used in many different scientific fields, which consider interactions between a living cell and a chemical(s). As a result, there are many definitions of varying detail and complexity in the scientific literature, which can lead to confusion and inconsistency."

For example, bioavailability can include (Semple et al. (2007):

- the portion of the total quantity or concentration of a chemical in the environment or a portion of it that is potentially available for biological action;
- the capacity of an organism's or a population's environment to provide a chemical relative to the capacity of the organism or population to transform that chemical;
- the amount of a compound taken up by an organism as the outcome of a dynamic equilibrium of organism-bound uptake processes and soil particle related exchange

[2] Many thanks to Dr. Bill Price for this example of differing bioavailability of mine waste within the human body.

processes, all in relation to a dynamic set of environmental conditions;
- the amount of chemicals in the soil that are present in forms that organisms can take up during the time they are living;
- the flux of contaminants to biota;
- the rate at which a chemical compound can be transported to the specific biological population;
- the degree to which chemicals present in the soil may be absorbed or metabolized by human or ecological receptors or are available for interaction with biological systems;
- what is accessible (as defined by mild extraction) and the contaminant's chemical activity (as defined by spontaneous physico-chemical processes, such as diffusion, sorption and partitioning);
- the maximum quantity of a contaminant available for uptake by an organism within a given time, which may describe two distinct fractions - bioavailability and bioaccessibility; and
- the degree to which chemicals present in the soil may be absorbed or metabolised by human or ecological receptors, or are available for interaction with biological systems.

What is the probability that the aqua-regia concentration of an element in a mine-waste sample reliably reflects and represents this true complexity of bioavailability? In most cases, the probability is vanishingly close to zero, and where it is representative there is no way to know in advance.

4. References

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