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WP 24-01

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GUIDANCE ON THE SELECTION OF DATA FOR THE DEVELOPMENT OF AQUATIC BIOACCUMULATION FACTORS

NCASI WHITE PAPER
WP 24-01
JANUARY 2024

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Acknowledgments
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Cite this report as:

NCASI. 2024. *GUIDANCE ON THE SELECTION OF DATA FOR THE DEVELOPMENT OF AQUATIC BIOACCUMULATION FACTORS*. White Paper (WP-24-01). Cary, NC: National Council for Air and Stream Improvement, Inc.

GUIDANCE ON THE SELECTION OF DATA FOR THE DEVELOPMENT OF AQUATIC BIOACCUMULATION FACTORS

EXECUTIVE SUMMARY

This white paper reviews best practices in conducting field studies to develop bioaccumulation factors (BAFs). This information will be of value to those who are reviewing field BAF studies and evaluating study quality to ensure that high quality data are relied on for the development of BAFs. This white paper will also be of value to those designing a protocol for the collection of data to derive a field BAF to ensure the study is robust and informative. This review covers the general approach to the derivation of field BAFs by US EPA and critical study elements that are needed in individual studies to ensure the use of robust and reliable data. Critical study elements include the number and timing of water samples, the number and timing of organism samples, the tissue target site of organism samples, the temporal coordination of water and organism samples, the spatial coordination of water and organism samples, the human health relevance of the species to which the BAF may be applied, the life history characteristics of the species to which the BAF may be applied (i.e., habitat preferences and food), and the characteristics of relevant non-water habitat (e.g., sediment chemical concentrations for benthic organisms or benthivores). Use of this guidance is intended to increase the transparency and reliability of field-derived BAFs for use in human health water quality criteria and other environmental risk assessment applications.

KEYWORDS

bioaccumulation factors (BAFs), fish tissue, trophic levels, biological accumulation, biological concentration, human health water quality criteria, persistent, bioaccumulative, toxic

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1.0 Introduction

New and existing substances are screened according to persistent, bioaccumulative, and toxic (PBT) pollutant criteria by regulatory agencies at the state, federal, and international level, such as the Stockholm Convention (UNEP 2001), Environment Canada (Environment Canada 2003), the US Environmental Protection Agency (EPA 1976), and the European Chemicals Agency (ECHA 2008). Substances classified as PBTs are potentially subject to regulatory controls beyond that of other chemical pollutants. The bioaccumulative potential of a chemical is a measure of the tendency for that chemical to form unacceptable concentrations in organisms through consumption of contaminated food or environmental exposure and is typically assessed by regulatory agencies through use of the n-octanol/water partition coefficient (K_{OW}), bioconcentration factor (BCF), and/or bioaccumulation factor (BAF) metrics. In general, threshold values of 1000 to 5000 are used for BCF and BAF, and values of 100,000 K_{OW} (Gobas et al. 2009). While many substances are classified as bioaccumulative based on laboratory BCF and BAF data and/or K_{OW} values, results from numerous field studies have shown widely differing levels of agreement between laboratory and field measurements of bioaccumulation potential of chemicals (e.g., Borgå et al. 2004; Weisbrod et al. 2009; Burkhard et al. 2012; Selck et al. 2012). For example, Arnot and Gobas (2006) noted that BAFs derived from field samples can differ by up to several orders of magnitude from laboratory-derived BCF measurements for some chemicals.

Laboratory studies encounter several specific areas of uncertainty that may contribute to inconsistencies between field- and laboratory-derived bioaccumulative estimates (NCASI 2022). For instance, the use of K_{OW} to predict bioaccumulative potential is suitable when the substance of concern is lipophilic and concentrates in the fat tissue of target species. However, when the substance of concern binds to protein or skeletal tissue, K_{OW} will not accurately predict bioaccumulative potential. Likewise, laboratory BAF derivation studies may provide an accurate characterization of the uptake and concentration potential of a substance dissolved in water at a specific concentration for aquatic organisms, but these studies fail to capture the effect of sediment loading and natural dietary habits, variation in environmental concentrations of the substance of concern, and other natural phenomena that affect consistency of the exposure pathway for relevant species.

Due to these challenges, US EPA explicitly prioritizes the use of field-derived BAFs for the development of national BAFs (EPA 2016). Field BAFs must be derived from carefully designed studies to ensure that concentrations measured in water bodies are relevant and predictive of the concentrations measured in aquatic organisms. This means study designs must consider the geographical location of sample collections, the sample size needed to capture concentration variability, lag time between water and organism sample collection, target tissue of organismal analysis, how to appropriately composite organismal samples, and how broadly representative selected organisms are, given the location and season. A study design that appropriately accounts for all of these factors is challenging to execute, and variations in these study design features can lead to orders of magnitude differences in field-derived BAFs. However, specific guidance on field-sampling approaches for the development of BAFs, and implications of deviating from best practices, is lacking.

Despite the regulatory priority for field-derived BAFs, from a study planning and execution perspective, they are challenging to derive and may be derived from incomplete or incoherent data. There is concern, from both a regulatory and industry perspective, of whether existing and newly derived BAFs accurately reflect actual bioaccumulation potential, resulting in either the overestimation or underestimation of the bioaccumulation potential of individual chemicals. Overestimation of bioaccumulation potential may lead to the unnecessary allocation of resources to further characterize a chemical hazard, whereas underestimation may lead to decisions that are not protective of environmental organisms or human health from the consumption of those organisms. This white paper reviews and

summarizes published guidance for selecting the highest quality data available for field-based BAF derivation to optimize and establish reliable field BAFs. The details provided in this paper will inform the development of field BAF studies to improve the characterization of bioaccumulative potential of substances in aquatic environments and ensure that subsequent water quality criteria reflect the best available data and methods.

2.0 General Field BAF Approach

US EPA provides a basic field BAF approach that is applicable to all chemicals (EPA 2003). The US EPA approach calls for a comparison of concentrations of the substance of concern in the water column/environment with the concentration of the substance in fish tissue, as shown in the following equation (EPA 2003, 2016). For lipophilic compounds, concentrations are adjusted for the lipid content of the tissue.

$$(\text{Baseline BAF})i = \text{BAF ffd Tt} - 1 \cdot f1\ell$$

where

$$(\text{Baseline BAF})i = \text{baseline BAF for field sample i (L/kg-lipid)}$$

BAFTt: total BAF from field sample (i.e., total concentration of chemical in tissue/total concentration of chemical in water [L/kg-tissue])

ffd: fraction of the total concentration of chemical in water that is freely dissolved

fℓ: fraction of tissue that is lipid

TL: trophic level

Multiple (Baseline BAF)i are averaged to a (Baseline BAF)TL n using the following procedure:

1. Compute the geometric mean across each chemical, computation method, TL, and species to compute a set of "species-mean baseline BAFs."

2. Compute the geometric mean across each chemical, computation method, and TL to compute a set of "trophic level-mean baseline BAFs" using the results from Step 1.

Additionally, when the accumulating tissue is not the lipid fraction of the organism, the equation should be adjusted to reflect this by using the whole-body mass of the organism or edible portion (muscle/fillet) if evaluating a trophic level relevant to human consumption.

While this approach outlines the basic components necessary for field BAF derivation, it does not address specific study design elements that ensure high data quality and a robust characterization of bioaccumulative potential. The following sections provide study design guidance that can be applied within the US EPA field BAF approach.

3.0 Critical Study Design Features

Sampling methods as well as sample processing and preparation for chemical analysis may affect measured concentrations in fish tissues and can contribute to variability seen in bioaccumulation endpoints. The guidance provided in this section is based on observations from critical reviews of BAF literature (Arnot and Gobas 2006, Burkhard

2003, 2021, Pignotti et al. 2017) as well as the primary sampling guidance provided by US EPA (EPA 2003). Critical features to evaluate in studies reporting BAFs and to consider when designing BAF development studies include:

- Number and timing of water samples.
- Number and timing of organism samples.
- Tissue target site of organism samples.
- Temporal coordination of water and organism samples.
- Spatial coordination of water and organism samples.
- Human health relevance of the species to which the BAF may be applied.
- Life history characteristics of the species to which the BAF may be applied (i.e., habitat preferences and food).
- Characteristics of relevant non-water habitat (e.g., sediment chemical concentrations for benthic organisms or benthivores).

The number of water samples should be sufficient to both modulate the variability of substance concentrations throughout the space of the water body and capture short-term variability in substance concentrations, such as those due to seasonality or rainfall. The number of samples needed will depend on the size and features of the body of water under study. Studies that rely on fewer than three water samples to characterize mean substance concentrations are considered insufficient for use in BAF development. Water bodies with more complex features may require more samples, both in terms of additional sample sites and additional sample replicates, to adequately relate mean water concentrations with aquatic organisms. Examples of water body characteristics necessitating additional water samples include geomorphological attributes that result in heterogeneous water quality conditions (such as inlets or backwaters) and vertical stratification of the water column in deeper lakes and river channels. Sediment loading can also be an important exposure pathway to characterize and may better capture all relevant inputs for bioaccumulation potential. Sediment samples may also be required for benthic species who feed near the bottom and may have sediment ingestion or experience higher water column concentrations near the sediment layer. Sediment samples should be paired with water samples to delineate the relative contribution of each pathway.

As indicated in Technical Bulletin 777 (NCASI 1999), chemical analyses of water, for the bioaccumulative chemical of interest and for any other parameter, should conform whenever possible to the US EPA-promulgated standard procedures in 40 CFR §136. An alternate reference may be the latest edition of *Standard Methods for the Examination of Water and Wastewater* (APHA 1995), which is a widely recognized source on analyses for water and wastewater and contains general guidance on water sampling. Methods contained in Test Methods for Evaluating Solid Waste (EPA 2023, EPA SW-846) may be appropriate under some circumstances. Draft methods should generally be avoided, particularly when method validation is incomplete. Water sampling considerations such as descriptions of and recommendations for collection equipment can be found in the *Handbook for Sampling and Sample Preservation of Water and Wastewater* (EPA 1982). Significant deviation from recommended data collection methods may impair study quality and the ability to reliably interpret study results.

In addition to water and sediment, sufficient organismal tissue samples should be collected to modulate variability from inter-individual dietary differences, variability due to movement, and differences related to life stage. The prioritization for tissue selection should reflect the trophic level to which the analysis is intended to apply. That is, if the organismal substance concentration concerns higher trophic level organisms that will consume the tissue, whole organism tissue composites should be used. For example, the substance concentration in benthic invertebrates should be measured in whole-body samples when they are expected to be consumed by benthivorous fish. When human consumption is the endpoint of concern, only edible portions of the organism should be analyzed (e.g., fillets). Even though substances may

partition in higher or lower concentrations to specific tissue components of aquatic organisms, the edible portion of tissue remains the most relevant exposure pathway to support risk assessment associated with bioaccumulative potential. Studies that rely on tissue components that exhibit higher bioaccumulative potential yet are inedible are likely to overestimate human exposure from the aquatic organism's bioaccumulative potential and are not optimal for BAF development. Specific guidance for predicting sample size requirements for field BAF studies can be found in Development of Site-Specific Bioaccumulation Factors (EPA 2009, Appendix 3A). Depending on the mass of the tissue sample required for analytical chemistry limits of detection, it may be necessary to composite tissue samples from multiple organisms of the same species. These composite samples should be from organisms sampled at the same time and location. Compositing tissue samples collected at different times and from different locations has the potential to introduce uncertainty into spatiotemporal tissue concentrations, and studies that composite samples in this way should be considered sub-optimal for BAF development.

There are no standard procedures promulgated by federal agencies on the handling and chemical analysis of fish tissue samples, but there are documents from US EPA and others that provide guidance on these matters. Among these are the Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories (EPA 1995). Options for fish and shellfish sampling methods are summarized in this reference, and recommended compositing procedures are also presented. This agency guidance also discusses various aspects of chemical analysis, including a list of general analytical techniques recommended for various analytes in fish and shellfish tissues. Another US EPA reference is the Assessment and Control of Bioconcentratable Contaminants in Surface Waters (EPA 1991), which contains procedural details for determining the presence and concentrations of bioconcentratable and bioaccumulative compounds. *Standard Methods for the Examination of Water and Wastewater* (APHA 1995) has general information on the acquisition of fish samples. Note that, generally speaking, chemical analysis of fish samples can be achieved using an analysis method from 40 CFR §136 intended for water. Sample preparation (extraction and cleanup) is not necessarily standardized, however. Significant deviation from accepted collection and preparation procedures may impair study quality and reliability.

Water and tissue samples should be spatially and temporally linked, with the temporal proximity of water concentration measures to those in biota dependent on the chemical characteristics of the compound of interest. Compounds with large K_{OW} will generally require multiple samples over time to establish the long-term chemical concentrations in the water, while substances with low K_{OW} require samples that are temporally proximate or concurrent to biota samples because the concentrations in tissue mimic those in water. For less hydrophobic chemicals, current chemical concentrations in the water provide a good predictor of the chemical concentration in tissue (EPA 2009). A disadvantage of field BAF studies compared to laboratory BCF studies is that field BAF studies cannot control for the movement of aquatic organisms in the body of water, which can introduce uncertainty into the water–tissue concentration relationship at a specific time and location. Although measured concentrations in aquatic organism tissues fundamentally relate to a previous exposure, the closer in time and space that water and tissue samples are collected increases the probability that water and tissue concentrations reflect a meaningful exposure pathway relationship.

If the toxicokinetic relationship between the chemical of concern and the organism is known, then an appropriate lag time between the water/sediment sample collection and organismal tissue collection can be developed. However, toxicokinetic data are often not available; therefore, study designs that collect tissue samples 1 to 2 weeks subsequent to water samples at the same location or water and tissue samples at the same time and location are optimal for establishing the water–tissue concentration relationship. As the spatial and temporal proximity between water and tissue sample collection increases, or if the concentration of high K_{OW} compounds is not reflective of average exposure concentrations, the less reliable data become for the development of a BAF. In general, data in which the time between water and tissue sampling is >1 year and the distance between water and tissue sample collection is >2 km are inadequate for BAF development (Burkhard 2021). However, under circumstances where water concentrations are

highly variable, where organism movement is highly variable, or where organismal metabolism and excretion is more rapid for the chemical of concern, data may need to have even shorter temporal lag times between water and tissue collection to be viable for BAF development.

The species selected for sampling should be relevant to the area for which the BAF will be applied; therefore, the location of sample collection should be relevant to the region for which it is intended to be applied. Aquatic species are adapted to the prevailing climate of the water body in which they live, which commutes a variety of physiological features that impact the bioaccumulative potential of substances to which they are exposed (e.g., water chemistry, habitat, flow, depth). For example, fish species who live in deep, cold bodies of water will be physiologically distinct from those who live in shallow, warm bodies of water in terms of lipid deposition, metabolic rate, feeding rate, sediment uptake rate, and potentially other adaptions that modulate bioaccumulative potential. As a result, field BAF studies that rely on species adapted to the region of concern will more accurately characterize the risk profile from the bioaccumulative potential of a substance, whereas applying a field BAF study conducted with species that are not regionally specific may substantially mischaracterize the risk from bioaccumulative potential.

4.0 Conclusions

Accurate BAFs are essential to the development of reasonable water quality standards that reflect true exposure scenarios and risk. However, BAFs are also heavily influenced by spatial and temporal sample collection decisions and sample processing methods. Currently, there are no explicit standards for data quality in the collection of substance concentrations in water bodies and aquatic organism tissue for the development of field BAFs. This is, in part, because the appropriate study design for generating BAFs that reflect true environmental conditions has not been evaluated. Also, the manner in which different study elements that affect BAF estimates has not been evaluated. This may account for the substantial differences in reported BAFs for similarly structured compounds, both within and across species. Critical study design and data quality components include the species selected for analyses, spatial and temporal proximity of biota and water samples, tissue type analyzed, sample size, and sample processing approaches. Generally, robust BAF studies are those that generate chemical concentrations from a sufficient number of environmentally representative water samples (n>3) and regionally relevant organism tissue samples that reflect the body portion eaten by the trophic level or consumer of interest. Additionally, collection of water and organism samples should be spatially and temporally proximate, ideally collected concurrently from the same location. When adequate sources of field BAF data are not available due to methodological issues with sample collection and analysis or regional relevance, it is advisable to consider primary data collection that relies on a robust approach for sampling, sample processing, and chemical analyses. A study that characterizes how BAFs and subsequent water quality criteria thresholds vary under different study design scenarios would fill an important information gap and improve the scientific basis of BAFs used for water quality criteria derivation.

References

Arnot, Jon A., and Frank A. P. C. Gobas. 2006. “A Review of Bioconcentration Factor (BCF) and Bioaccumulation Factor (BAF) Assessments for Organic Chemicals in Aquatic Organisms.” *Environmental Reviews* 14, no. 4: 257–97. https://doi.org/10.1139/a06-005.

APHA (American Public Health Association). 1995. *Standard Methods for the Examination of Water and Wastewater.* 19th Edition. Washington, DC: American Public Health Association.

Borgå, Katrine, Aaron T. Fisk, Paul E. Hoekstra, and Derek C. G. Muir. 2004. "Biological and Chemical Factors of Importance in the Bioaccumulation and Trophic Transfer of Persistent Organochlorine Contaminants in Arctic Marine Food Webs." *Environmental Toxicology & Chemistry* 23, no. 10: 2367–85. https://doi.org/10.1897/03-518.

Burkhard, Lawrence P. 2003. "Factors Influencing the Design of Bioaccumulation Factor and Biota-Sediment Accumulation Factor Field Studies." *Environmental Toxicology & Chemistry* 22, no. 2: 351–60. https://doi.org/10.1002/etc.5620220216.

———. 2021. "Evaluation of Published Bioconcentration Factor (BCF) and Bioaccumulation Factor (BAF) Data for per- and Polyfluoroalkyl Substances Across Aquatic Species." *Environmental Toxicology & Chemistry* 40, no. 6: 1530–43. https://doi.org/10.1002/etc.5010.

Burkhard, Lawrence P., Jon A. Arnot, Michelle R. Embry, Kevin J. Farley, Robert A. Hoke, Masaru Kitano, Heather A. Leslie, Guilherme R. Lotufo, Thomas F. Parkerton, Keith G. Sappington, Gregg T. Tomy, and Kent B. Woodburn. 2012. "Comparing Laboratory and Field Measured Bioaccumulation Endpoints." *Integrated Environmental Assessment & Management* 8, no. 1: 17–31. https://doi.org/10.1002/ieam.260.

Environment Canada. 2003. "Guidance Manual for the Categorization of Organic and Inorganic Substances in the Canada's Domestic Substances List, Determining the Persistence, Bioaccumulation and Inherent Toxicity," Accessed January 19 2011. http://www.ecgc.ca/Substances/ese/eng/dsl/guidance_document.cfm.

ECHA (European Chemicals Agency). 2008. Chapter R.11. "PBT Assessment." In *Guidance on Information Requirements and Chemical Safety Assessment*. Helsinki, Finland: European Chemicals Agency.

Gobas, Frank A. P. C., Watze de Wolf, Lawrence P. Burkhard, Eric Verbruggen, and Kathleen Plotzke. 2009. "Revisiting Bioaccumulation Criteria for POPs and PBT Assessments." *Integrated Environmental Assessment & Management* 5, no. 4: 624–37. https://doi.org/10.1897/IEAM_2008-089.1.

NCASI (National Council for Air and Stream Improvement, Inc.) 1999. Guidance on the Site-Specific Evaluation of Bioaccumulation Factors Under the Great Lakes Water Quality Initiative. Technical Bulletin No. 777. Research Triangle Park, NC.

NCASI (National Council for Air and Stream Improvement, Inc.) 2022. Bioaccumulation Potential of PFAS in Fish. Technical Bulletin No. 1080. Research Triangle Park, NC.

Pignotti, Emanuela, Gemma Casas, Marta Llorca, Anil Tellbüscher, David Almeida, Enrico Dinelli, Marinella Farré, and Damià Barceló. 2017. "Seasonal Variations in the Occurrence of Perfluoroalkyl Substances in Water, Sediment and Fish Samples from Ebro Delta (Catalonia, Spain)." *Science of the Total Environment* 607–608: 933–43. https://doi.org/10.1016/j.scitotenv.2017.07.025.

Selck, Henriette, Ken Drouillard, Karen Eisenreich, Albert A. Koelmans, Annemette Palmqvist, Anders Ruus, Daniel Salvito, Irv Schultz, Robin Stewart, Annie Weisbrod, Nico W. van den Brink, and Martine van den Heuvel-Greve. 2012. "Explaining Variability of Bioaccumulation Measurements Between Laboratory and Field Using a Modelling Approach." *Integrated Environmental Assessment & Management* 8, no. 1: 42–63. https://doi.org/10.1002/ieam.217.

UNEP (UN Environment Programme). 2001. Stockholm Convention on Persistent Organic Pollutants, Annex D. http://www.pops.int.

USEPA (US Environmental Protection Agency). 1976. Toxic Substance Control 1 Act (TSCA). 15 US Code 2601 et Seq. Washington, D.C.

———. 1982. Handbook for Sampling and Sample Preservation of Water and Wastewater. EPA 600/4-82/029. Washington, D.C.

———. 1991. Assessment and Control of Bioconcentratable Contaminants in Surface Waters. EPA 833-D94-001. Washington, D.C.

———. 1995. Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories. Volume I: Fish Sampling and Analysis, 2nd Edition. EPA 823-R-95-007. Washington, D.C.

———. 2003. Methodology for Deriving Ambient Water Quality Criteria for the Protection of Human Health (2000), Technical Support Document. Volume II: Development of National Bioaccumulation Factors. EPA 822-R-03-030. Washington, D.C.

———. 2009. Methodology for Deriving Ambient Water Quality Criteria for the Protection of Human Health (2000), Technical Support Document. Volume III: Development of Site-Specific Bioaccumulation Factors. EPA 822-R-09-008. Washington, D.C.

———. 2016. Development of National Bioaccumulation Factors: Supplemental Information for EPA's 2015 Human Health Criteria Update. EPA 822-R-16-001. Washington, D.C.

———. 2023. Test Methods for Evaluating Solid Wastes. EPA SW-846. (Last updated November 17, 2023). https://www.epa.gov/hw-sw846.

Weisbrod, Annie V., Kent B. Woodburn, Albert A. Koelmans, Thomas F. Parkerton, Anne E. McElroy, and Katrine Borgå. 2009. "Evaluation of Bioaccumulation Using In Vivo Laboratory and Field Studies." *Integrated Environmental Assessment & Management* 5, no. 4: 598–623. https://doi.org/10.1897/IEAM_2009-004.1.