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Brief Communication

Progress in the environmental risk assessment of plant protection products in Brazil: An overview of aquatic organism proposals

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Abstract

Since 2019, the Brazilian Institute of Environment and Renewable Natural Resources (IBAMA) has actively developed pesticide environmental risk assessment (ERA) frameworks adapted to Brazil's specific ecological contexts. This endeavor, supported by funding from the Brazilian Ministry of Justice and in partnership with academic institutions, has led to a concerted effort to establish ERA protocols for various taxa, including birds and mammals, soil organisms, aquatic organisms, and reptiles and amphibians. The outcomes of this initiative were conveyed in two workshops held in February and November 2023, during which the agency communicated its findings to the technical-regulatory community. This article represents one of two articles that provide more detailed insights into the ERA propositions for all taxa. In this article, we summarize the proposals for aquatic organisms presented and discussed during the workshops, which can be used as an informational source by the technical-regulatory community. *Integr Environ Assess Manag* 2024;20:1787–1792. © 2024 The Author(s). *Integrated Environmental Assessment and Management* published by Wiley Periodicals LLC on behalf of Society of Environmental Toxicology & Chemistry (SETAC).

KEYWORDS: Aquatic invertebrates; Aquatic plants; Environmental risk assessment; Freshwater fish; Regulatory paradigm change

INTRODUCTION

Since 2019, the Brazilian Institute of Environment and Renewable Natural Resources (IBAMA) has initiated discussions on the implementation of pesticide environmental risk assessment (ERA) guidelines for different taxa such as birds and mammals, soil organisms, aquatic organisms, and reptiles and amphibians (Diário Oficial da União Official Gazzette [DOU], 2019). These discussions were preceded by earlier ones about aquatic organisms and pollinators. In the latter case, the first risk assessment guideline was published in 2017 (Cham et al., 2017). In 2019, the Ministry of Justice funded the project, a collaborative effort between IBAMA and academia (Instituto Brasileiro do Meio Ambiente [IBAMA], 2020, 2021), pooling their knowledge and expertise to identify and address knowledge gaps (e.g., the need to incorporate new studies, especially with local tropical species) to promote a discussion of how to estimate the exposure of nontarget organisms to plant protection products (PPPs) and to evaluate current requirements regarding ecotoxicological studies. The results of these efforts were disseminated in two workshops in 2023 (Instituto Brasileiro do Meio Ambiente [IBAMA], 2023a, 2023b), where the findings until that moment were shared with the technical-regulatory community. In parallel, on 27 December 2023, a new paradigm in pesticide registration in Brazil was approved by the Brazilian Congress (Brasil, 2023), where the pesticide dossier evaluation will now progress from a hazard-based classification system to a risk assessment procedure, which became mandatory. Cione et al. (2024) published a more detailed discussion of this subject.

This article summarizes the changes presented by IBAMA at a two-day workshop held in November 2023. It is one publication in a series of two that focus on the ERA proposals for aquatic organisms (Instituto Brasileiro do Meio Ambiente [IBAMA], 2023c). In this article, the authors intend to share the information presented by IBAMA during the workshop with local and global stakeholders (pesticide industry, risk assessors, academics, researchers, and others) because the guidance and supporting documents from the agency are not yet available. The workshop was recorded; therefore, the original information presented by IBAMA is accessible on YouTube videos in Portuguese (IBAMA, 2023c). It is essential to mention that this article is a snapshot of the workshop in which IBAMA continues to evolve in the implementation process. To convey the full message articulated by IBAMA, a similar article on birds, mammals, and soil organisms is on page 1793.

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AQUATIC ORGANISMS: RECENT UPDATES OF EXPOSURE AND ECOTOXICOLOGICAL EFFECTS

The second ERA workshop organized by IBAMA in November 2023 (IBAMA, 2023a) focused primarily on the discussion of the ecotoxicological studies, including testing with local native species, species sensitivity distribution (SSD), and the identification of knowledge gaps (e.g., studies with new species under adapted local guidelines). Conversely, aspects of the aquatic risk assessment exposure component were addressed broadly during the first workshop in February 2023 (IBAMA, 2023b). As presented by the agency, four technical documents were published, focusing on the following themes:

(a) New requirements to assess the exposure and effect of pesticides on aquatic organisms aligned with actual legal requirements in Portaria 84 (Instituto Brasileiro do Meio Ambiente [IBAMA] e dos Recursos Naturais Renováveis, 1996);
(b) Additional extrapolation factors to be considered during the initial stages of aquatic ERA, accounting for the differences between Brazilian native species;
(c) Recommendations for free software tools to build SSD curves;
(d) Guidance on conducting, assessing, and interpreting results from microcosm and/or mesocosm studies—the final document summarizes recommendations from the Organisation for Economic Co-operation and Development (OECD, 2006a) and recommendations from the working group of the Diffuse Right Fund (FDD in Portuguese) task force (Instituto Brasileiro do Meio Ambiente [IBAMA], 2021).

In subsequent sections, we highlight the findings communicated in the second workshop, which focused primarily on the results of the first two technical documents. Nonetheless, in the text, we have summarized proposals on the exposure of aquatic organisms to pesticides for elucidation purposes (IBAMA, 2023b).

Exposure: Proposals for an aquatic exposure tool and development of local exposure scenarios

Exposure of aquatic organisms to PPPs was discussed during the first workshop convened by IBAMA in February 2023 (IBAMA, 2023c). In this workshop, IBAMA named the USEPA Pesticide in Water Calculator (PWC; USEPA, 2024) as the designated exposure tool for estimating the predicted environmental concentration in surface water (PEC_{SW}) for aquatic organism risk assessment in Brazil (IBAMA, 2023a). Additionally, it is essential to highlight that, because of collaborative efforts involving EMBRAPA and IBAMA, 19 distinct local crop-specific scenarios were developed for use in the PWC tool; these scenarios were released by IBAMA in April 2024 (Instituto Brasileiro do Meio Ambiente [IBAMA], 2024). Moreover, EMBRAPA and CropLife Brasil (DOU, 2024) recently published a collaboration term to develop additional crop-specific scenarios and propose how mitigation measures can be used in the PWC tool for some scenarios. The agency also aims to identify the most important input parameters for applying the PWC in Brazilian regulatory decisions.

Effects

(a) *Building a local database, including standard and local tropical species*
One initiative of the FDD project was a literature review to compare ecotoxicological endpoints between tropical and temperate species. Local academics conducted this study and performed a comprehensive literature search on aquatic ecotoxicity; the survey results revealed 522 bibliographic references, of which 327 were concerned with tropical and 195 with temperate species. It was reported that the review of these manuscripts identified endpoints for 87 active ingredients, encompassing data for 115 tropical and 98 temperate species. As stated during the workshop, this database will help define new study requirements and extrapolation factors for Tier 1.
(b) *Updates on ecotoxicological studies and comparison between Brazilian and foreign regulatory requirements*
During the workshop, one main discussion centered on ecotoxicological studies of aquatic organisms, and the need to update current national guidelines was pointed out (IBAMA e dos Recursos Naturais Renováveis, 1996). The deliberations also involved an extensive comparative analysis of Brazilian regulatory requirements with those of foreign agencies, namely the USEPA and European Food Safety Authority (EFSA), focusing specifically on the ecotoxicity parameters assessed for each trophic level and their responses to the pesticides under evaluation. A synthesis of the differences among national, USEPA, and EFSA requirements for ecotoxicological testing is shown in Table 1.During the presentation, details on how effects for each taxon are measured and how the endpoints are derived were presented, highlighting the differences between local requirements and those of the USEPA and EFSA. Nonetheless, because some of the principles presented constitute basic ecotoxicological principles and considering that the focus of this article is to report on the workshop's main findings, they were not transcribed in this article. However, they can be found in each agency's guidance documents (European Food Safety Authority [EFSA], 2013; USEPA, 2004) to justify testing species requirements with the mode of action of pesticides; for example, for insecticides, the necessity was recognized for chronic testing of *Chironomus* sp. when the substance accumulates in sediment or when the pesticide is known to interfere with molting hormones. Regarding fish, as indicated in Table 1, tests can be performed at early life stages or for full life cycles. As for algae, upon comparing the data required by the USEPA and EFSA, IBAMA observed that both agencies require the green algae *Raphidocelis subcapitata*; therefore, it will potentially be

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TABLE 1 Comparison between the ecotoxicological parameters adopted by IBAMA and those adopted by the USEPA and EFSA[a]

Nontarget organisms	IBAMA e dos Recursos Naturais Renováveis (1996)	USEPA (2004)	EFSA (2013)[b]
Acute tests			
1. Aquatic invertebrates: Cladocera	–	*Daphnia magna* or other more sensitive aquatic invertebrate species	*Daphnia* sp. (all types of pesticides)
2. Aquatic Invertebrates: others	Tests with microcrustaceans	–	Additional arthropod species (insecticides only)
3. Fish	Tests with fish	Most sensitive of the following suggested species: *Lepomis macrochirus*, *Oncorhynchus mykiss*, and *Pimephales promelas*	*Oncorhynchus mykiss* (all types of pesticides)
Chronic tests			
1. Algae	Tests with algae	Five species are suggested, where the most sensitive is chosen (including the aquatic plant species): *Skeletonema costatum*, *Anabaena flos-aquae*, *Raphidocelis subcapitata*, *Chlorella vulgaris*, and *Desmodesmus subspicatus*	Herbicides: one species of green algae and one species of nongreen algae Insecticides and other pesticide classes: one species of green algae
2. Aquatic plants	–	*Lemna gibba* (if more sensitive than algae)	The most suitable species among the following options: *Lemna* sp., *Myriophyllum* sp., or *Glyceria maxima* (herbicides)
3. Aquatic invertebrates: Cladocera	–	*Daphnia magna* or other more sensitive aquatic invertebrate	Insecticides: *Daphnia* sp. or other more sensitive arthropods Herbicides and other pesticide types: *Daphnia* sp.
4. Aquatic invertebrates: others	Tests with microcrustaceans	–	Insecticides: *Chironomus* sp. in exceptional cases Herbicides and other pesticide types: *Chironomus* sp. or *Lumbriculus* sp. in exceptional cases
5. Fish	Tests with fish Bioconcentration studies with fish (triggered by specific conditions)	Most sensitive of the following suggested species: *Lepomis macrochirus*, *Oncorhynchus mykiss*, and *Pimephales promelas*	All pesticide classes: initial life stages or full life cycle Bioconcentration studies (in specific cases)

Abbreviations: EFSA, European Food Safety Authority; IBAMA, Brazilian Institute of Environment and Renewable Natural Resources.
[a]Transcribed as presented by IBAMA in the second workshop on the scientific–technical basis of pesticide environmental risk assessment (IBAMA, 2023c).
[b]In the presentation, IBAMA indicated that, in the EFSA guidance document (2013), the tests are described considering different classes of pesticides; therefore, the specificity of each test is highlighted.

included as a regulatory requirement in Brazil when new regulations are published (see Table 2).

For herbicides, if *Lemna* and other algae species are not sensitive to the substance, a rooted macrophyte may be required (preferably *Myriophyllum* sp.). The agency further indicated that tests with *Glyceria maxima* may be required when data suggest terrestrial plants (e.g., monocotyledons) are also susceptible to herbicides. As indicated by IBAMA, chronic tests with *Chironomus* sp. and *Lumbriculus* sp. may be required if the substance is identified as accumulating in sediment. Additional requirements for herbicides and other pesticides are listed in Table 1. In all cases, the acute endpoints are EC50 and LC50; in chronic studies, the endpoints are EC10, NOEC, and ECr (for algae growth rate).

CONSIDERATION OF LOCAL TROPICAL SPECIES VERSUS STANDARD GUIDELINE SPECIES

As indicated in Table 1, during the workshop, the absence of some nontarget organisms—compared with the USEPA and EFSA guidelines—was noted in the Brazilian requirements; for instance, there is no reference to Cladocerans for

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TABLE 2 Description of suggested species for adoption as biological models in aquatic organisms ERA[a]

Group	Species	Main endpoints	Normative
Primary producers, algae	*Raphidocelis subcapitata*	Algal growth	NBR 12648 (ABNT, 2018)
Primary producers, aquatic plants	*Azolla caroliniana* and *Salvinia molesta*	Fresh and dry biomass, foliar area, root length	Adaptation of OECD 221 (Organisation for Economic Co-operation and Development [OECD], 2006b)
Invertebrates, Cladocera	*Ceriodaphnia silvestrii*	Immobility (acute test) and reproduction (chronic test)	NBR 12713 (Associação Brasileira de Normas Técnicas [ABNT], 2022b) and NBR 13373 (Associação Brasileira de Normas Técnicas [ABNT], 2017)
Invertebrates, Insecta	*Chironomus sancticaroli*	Mortality (acute test), survival and growth (subchronic test), survival, growth, emergence, and development (chronic test)	Adaptation: Fonseca and Rocha (2004), OECD 218 (OECD, 2004), and OECD 233 (OECD, 2011)
Invertebrates, Amphipoda	*Hyalella* sp.	Lethality (acute test), survival and growth (chronic test)	NBR 15470 (ABNT, 2013)

Abbreviations: ERA, environmental risk assessment; IBAMA, Brazilian Institute of Environment and Renewable Natural Resources.
[a]As presented by IBAMA in the second ERA workshop, aquatic organisms section (IBAMA, 2023b).

both acute and chronic tests, whereas for aquatic plants, chronic studies do not refer to tests with macrophytes. The results of ecotoxicological tests using native Brazilian species were presented by IBAMA.

Primary producers

In the ecotoxicological database built during this project, out of 76 studies performed on algae, 66 were performed on *Raphidocelis subcapitata*. Therefore, as previously explained, IBAMA indicated that these species may be used for testing of primary producers. Additionally, there are local guidance documents to perform tests with these species (NBR 12648; Associação Brasileira de Normas Técnicas [ABNT], 2018), which include not only this species of algae but additional algae (*Chlorella vulgaris*, *Scenedesmus subspicatus*, *Monoraphidium dybowskii*). Therefore, this reinforces the proposal to include *R. subcapitata* in the national requirements.

Regarding macrophytes, IBAMA reported that seven studies with four native species were found: *Azolla caroliniana*, *Pistia stratiotes*, *Salvinia molesta*, and *Wolffia brasiliensis*. The studies were conducted by adapting the OECD guidelines for *Lemna* sp., which evaluates plant growth (dry and fresh biomass, number of fronds, leaf area, and root size). The importance of studies of macrophytes was noted by IBAMA because they are more sensitive to certain herbicides (Cedergreen & Streibig, 2005).

Aquatic invertebrates

(a) *Cladocerans*

As previously mentioned, Brazilian legislation does not indicate any tests for Cladocerans, unlike the USEPA and EFSA, which require data for *Daphnia magna* or more sensitive species (Table 1). In IBAMA's database, studies were found for seven native species: *Ceriodaphnia silvestrii*, *Ceriodaphnia rigaudi*, *Daphnia laevis*, *Moina minuta*, *Pseudosida ramosa*, *Diaphanosoma birgei*, and *Bosmina freyi*, where data for *C. silvestrii* comprise 10 of 16 evaluated studies, yielding 127 of 141 endpoints. Based on an SSD curve performed using carbofuran, cypermethrin, and chlorpyrifos, IBAMA highlighted the sensitivity of *C. silvestrii* compared with other taxonomic groups. Species sensitivity distribution curves obtained using 2,4-D, atrazine, and glyphosate also revealed the high sensitivity of *C. silvestrii*, which is the species most sensitive to atrazine. Although not shown, this aquatic invertebrate was also the species most sensitive in SSD curves for fipronil, ametryne, and diuron. Finally, to justify the inclusion of *C. silvestrii* in national requirements IBAMA showed a national ecotoxicological guideline available for this species (NBR 13373; ABNT, 2017), including acute and chronic tests.

(b) *Other invertebrates*

In the presentation, it was noted that both the USEPA and the EU consider other invertebrate groups in addition to daphnids, whereas in Brazil, they are required (Table 1). It was mentioned that testing other groups, such as Insecta, Oligochaeta, Amphipoda, and Ostracoda, is important. For Insecta, IBAMA noted that *Chironomus sancticaroli* has been used most often and that literature reviews demonstrate that tests with species are viable with other substances in addition to pesticides. Additionally, the adaptation of international protocols was deemed satisfactory (Organisation for Economic Co-operation and Development [OECD], 2004, 2011). Regarding laboratory cultivation, Fonseca and Rocha (2004) described a protocol for *C. sancticaroli*, in which the endpoints were mortality in acute tests, survival and growth (size and biomass) in subchronic tests, and full life cycle for the species considering survival, growth, emergence, and development. It was also demonstrated that

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C. sancticaroli has a shorter life cycle than other standard species such as *C. riparius* and *C. tentans* (Pinto et al., 2021).

The presentation also indicated that studies of amphipod species (*Hyalella* sp.) were found in IBAMA's database. Species sensitivity distribution curves were built using acute endpoints for native species—*H. meinert*, *H. curvispina*, *H. castroi*, and *H. pleocuta*, with *H. meinert* the species most sensitive in SSD curves performed with fipronil. Additionally, it was noted that there are national ecotoxicological guidelines for both acute and chronic testing of *Hyalella* sp. (Associação Brasileira de Normas Técnicas [ABNT], 2013). It was also mentioned that the standard species *H. azteca*, widely used globally, is not registered in the Brazilian Biodiversity Information System (https://www.sibbr.gov.br/).

Fish

As indicated by IBAMA, fish is the taxon with the most significant volume of data available, with 124 studies covering 1086 toxicity data. These studies consider lethal and sublethal effects, behavior, growth and reproduction, biochemical biomarkers, genetic responses, and histology. Of the 33 species, six orders were identified: Characiformes, Cichliformes, Cyprinodontiformes, Gymnotiformes, Siluriformes, and Synbranchiformes. For fish, it was difficult to establish a sensibility scale due to the wide range of responses—from the individual (e.g., growth) up to populational (e.g., reproduction). However, IBAMA stated that, when applying SSD for herbicides and insecticides to the data, three species were more often identified: *Rhamdia quelen*, *Cnesterodon decemmaculatus*, and *Prochilodus lineatus*. National guidelines are available (Associação Brasileira de Normas Técnicas [ABNT], 2022a) for the ecotoxicological testing of *Danio rerio* and *Pimephales promelas*.

Proposals for ecotoxicological testing, including local native species

After summarizing the workshop's discussion of ecotoxicological testing, comparing the different requirements between Brazil and other regions, and documenting the availability of endpoints with local native species, IBAMA presented a proposal that called for the species to be representative and referred to their respective testing guidelines, which are summarized in Table 2.

FINAL REMARKS

This article summarizes highlights of the most recent developments in the aquatic ecotoxicology of pesticides during the second workshop on pesticide ERA in Brazil. The authors demonstrate the evolution of this topic up to the present, which may contribute to discussions regarding recent regulatory paradigm changes, as indicated by the enactment of Law 14.785/2023 (Brasil, 2023; Cione et al., 2024), which has declared pesticide risk assessment mandatory in Brazil. This achievement aligns Brazil more closely with the regulatory science standards of developed nations and some neighboring countries such as Colombia, Ecuador, and Peru (Secretaria General de la Comunidad Andina [SGCAN], 2002).

AUTHOR CONTRIBUTIONS

Ana Paola Cione: Conceptualization; data curation; formal analysis; project administration; supervision; writing—original draft; writing—review and editing. **Gustavo Souza Santos**: Writing—original draft; writing—review and editing. **Fábio Casallanovo**: Visualization; writing—original draft; writing—review and editing.

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CONFLICT OF INTEREST

All authors declare that the Syngenta companies employ them, as declared in their affiliations.

DISCLAIMER

The peer review for this article was managed by the Editorial Board without the involvement of Ana Paola Cione, Gustavo Souza Santos, and Fábio Casallanovo.

DATA AVAILABILITY STATEMENT

Because this brief communication conveys a summary from a workshop, no experimental data are provided. The sources of information are mentioned in the References section.

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