



Amphibians as Bio-indicators of Environmental Health

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Abstract

Amphibians function as reliable bio-indicators of environmental health by integrating multi-stressor signals, reflecting habitat integrity, water quality, and ecosystem function through physiological, genetic, and population-level responses. Amphibians are effective bio-indicators of environmental health due to their ecology and functional traits. Bio-indicators are organisms whose responses to environmental changes reliably reflect broader ecological conditions. Sensitivity indicates how a taxon reacts to stressors, while exposure refers to the stressor's nature, intensity, and duration. The terms effect and response describe observable changes in physiology, life-history traits, behavior, or community structure due to exposure. Amphibians are impacted by various stressors, including chemical contaminants, temperature shifts, and habitat changes, making them ideal bio-indicators. Other organisms, like fish and invertebrates, can also be studied, but amphibians effectively address a wide range of stressors with notable sensitivity.

Amphibians, among the most at-risk vertebrates, face numerous stressors, including pollution, climate change, habitat loss, and pathogens. Key chemical contaminants of concern are pesticides, heavy metals, herbicides, and pharmaceutical residues. Climatic extremes like drought and flooding have noticeable effects. Changes in hydroperiod impact breeding opportunities, while temperature influences developmental timing, affecting fitness and population viability. Habitat integrity is crucial, with land use and water quality affecting long-term sustainability. Pathogens like chytridiomycosis and ranaviruses pose growing threats. Studying amphibians can provide valuable insights into the impacts of various stressors. (Soto-Rojas et al., 2017)

Keywords: Amphibians, environmental health, bio-indicators.

1. Introduction

The remarkable loss of biodiversity observed globally is attributed to the proliferation of various stressors while reduced financial resources and scientific manpower challenge conservation activities. To prioritize animals or habitats that require conservation, indicators of environmental disruptions have been utilized. An indicator is defined as a variable that translates information from complex systems into simpler, manageable information multiple at the same time, in a form intuitive to humans and likely to trigger concern, and for which direct measurements are inexpensive and can be made frequently. Among the various potential indicators, amphibians are increasingly regarded as the vehicle integrating multiple pressures on the environment and their condition is believed to reflect the integrity of environmental components and functions since most of the amphibians' lifetime is aquatic while the adult period is terrestrial. Consequently, variations of bio-indicator theory had been applied to amphibians, as they are sound candidates at individual and population levels to monitor the general changes of the ecosystem. Attention to physiological variation that change depending on the type of environmental perturbation suffered had been induced. Very recently the decrease of water bodies and declines in hydroperiod had been introduced as additional factors affecting the monitoring process reflected by amphibians and seedlings used from a watershed scale approach at the species level. Amphibians are especially sensitive to pollutants. Pollutants loading, known from human population increase, lands reduction, and forest degradation should be monitored in order to safeguard amphibians. On the other hand, as global warming accelerates the oscillation of temperature and wet meter cycle species and life stages in the tropics and sub-tropics connected with eco-physiology must be closely considered when designing global warming projects. A large range of diseases have been observed at various aquatic stages and for those in temporary or lotic water bodies with high water flow preparing sub-criteria are useful (Strong et al., 2017).

2. Conceptual Framework: Bio-indicators and Amphibians

Amphibians represent an invaluable tool for assessing ecosystem environmental health by indicating the presence of correct aquatic, temporal, and multiple stressors in their life-cycle. Bio-indicators are species or assemblage that reflect habitat quality through presence or abundance. Sensitive species fulfill integration capacity gaps for such stressors. Bio-indicator frameworks establish indicator hierarchies for tracking pathways and responses across scales that guide environmental monitoring objectives. Anthropogenic changes act as “active” stressors like chemical runoff or “passive” stressors like nutrient enrichment, micro-structure, and wetness. Active and passive

changes trigger different species assemblage shifts and provide complementary monitoring. After years of study, attitudes towards pond-bleaching must fundamentally shift. The absence of taxa like Pelobates or Planobates swamp the remaining glacial viridis complex assemblages, providing reinforcement of presence as indication they are “never-pool” (Hadi Fikri et al., 2013).

Different land-use patterns alter water quality in sedimentary regions between within river basins. Changes in measureable water-quality parameters had since well-established links for species assemblages and broader taxonomic groups, ensuring sampling effort and primary focus remain on community realizations, micro-hydrology, and qualitative descriptions (Strong et al., 2017).

3. Amphibian Sensitivity to Environmental Stressors

Amphibians play a crucial role as bio-indicators of environmental health. Their amphibious life-cycle and dependence on aquatic habitats make them sensitive to multiple stressors and, at the same time, integrators of contaminants from different sources and land-use patterns. All these qualities make this group very suitable for assessment of ecosystem health. The absence of amphibians from a landscape can indicate grave problems such as habitat destruction and/or degradation.

Amphibians can detect stressors ranging from chemical contamination and climate alteration to habitat modification and disease introduction. Such pressures can lead to physiological and immunological mis-regulation, chronic stress, alterations in reproductive traits such as male to female ratio and timing of metamorphosis, poor health condition, significant shifts in species assemblage, and ultimately to biotic collapse of communities. These responses-integration across various levels of biological organization and linkage to diverse environmental perturbations-greatly favour the application of amphibians as reliable bio-indicators.

Chemoclines within aquatic systems can point the sources of contamination and/or the exogenous factors affecting a specific territory. These reliable indicators are both constituents of Global Champions 99, although they are also recorded in Caribbean terrestrial systems seldom integrated in global programmes.

4. Methodologies for Monitoring Amphibian Health and Habitat Quality

Amphibian health and the quality of aquatic and terrestrial environments can be monitored through a variety of methodologies that utilize multi-level physiological, immunological, genetic, and molecular metrics. The main types of monitoring are field-based methods that assess amphibian occupancy, abundance, and calling activity, along with estimates of habitat quality, and laboratory techniques that measure stress-induced

physiological responses, immune system performance, population health, gene expression, pathogen presence, and genetic diversity. Monitoring goals and the corresponding metrics and data streams of interest are defined.

Field surveys of amphibian occupancy, abundance, calling activity, and habitat quality that capture the temporal and spatial scales of interest can be conducted by trained personnel along sites selected to represent chapter-3 monitoring strata. Sufficient replication is achieved through the allocation of predetermined sampling effort, which is determined for each indicator based on site availability and the prevailing feasibility-conservation trade-offs. Two types of methods are applied independently, each providing a complementary means of assessing the health of amphibians and the habitats they depend upon. One method targets individual or community capacity to accumulate, tolerate, and recover from disturbances along a an explicit gradient. Multi-marker indices summarize multi-parametric data to deliver a single integrated measure of environmental health.

Laboratory assessments that evaluate physiological, immunological, genetic, and molecular responses to stress, disease, and environmental quality are accompanied by detailed sampling protocols for both wild and captive specimens to facilitate inter-study comparability. Several stress-related biomarkers whose variations communicate exposure or sensitivity under given stressor regimes are routinely measured and subsequently interpreted through an explicitly defined context-dependent framework that accommodates compensatory adjustments, regime shifts, and long-term trends. Monitoring programs in various socio-ecological contexts rely on temporal and spatial data describing hazardous substances, basic habitat characteristics, and relevant biotic composition, which serve to contextualize temporal trends in ambient hazard exposure, population or community impairment, and habitat alteration.

The assessment of amphibian health, populations, community composition, and the quality of aquatic and riparian habitats encountered by aquatic-breeding amphibians can be accomplished throughout the amphibian lifecycle—before, during, and after aquatic activity—by measuring the activity of certain biomarkers under a defined context. Stress-induced hormone levels changing due to the activity of chemicals such as pesticides, heavy metals, hydrocarbons, hydrocarbons, or yet-to-be-identified redirection drugs can be monitored along the temporal, estacion, and visa-sight variations. The measurement of these biomarkers (which can be obtained in the field) leads to a holistic understanding of the landscapes encountered, thus enabling a clearer representation of the health of the entire system.

4.1. Field Survey Techniques

Amphibians are incorporated through a range of widely employed monitoring protocols or techniques. Presence–absence and occupancy models (robust design, single season, multi-season) inform species distribution patterns, habitat associations, and demographic trends; abundance indices based on call surveys assist landscape assessments; time-constrained visual encounter surveys cover broader species assemblages; and fragment occupancy studies quantify critical threshold densities and habitat use by the range-restricted species *Oedipina* spp. Targeted investigations in seasons of species activity accumulate pertinent data on community composition, functional structure, and gene flow. These techniques support sampling consistent with distinct geographic areas—forest-grown freshwater wetlands, cool-temperate forest springs and headwater patches, and urban–rural riverscape gradients in rural southeastern Australia, as well as temperate–tropical agricultural–urban landscapes in southern Queensland.

Field-based survey protocols can be adjusted to estimate local amphibian occupancy or abundance, especially for high-calling taxa, with species detection history integrated into national databases. Temporal and spatial variability in amphibian calling and detection require careful consideration of sampling effort and replication, and these issues are particularly acute for phylogenetically nested or closely allied species. Time-constrained Visual Encounter Surveys (VES) in the nonbreeding season offer another approach for assessing wide ranges of species within specific area discontinuity, although they may not adequately represent habitat-specific distribution patterns.

4.2. Laboratory Assessments

Monitoring amphibian health and habitat quality employs several methodologies to evaluate overall ecosystem functionality. Specific monitoring goals define the associated metrics and data streams. Field surveys have proven most effective at assessing amphibian health in the field. Monitoring amphibian occupancy, abundance, and calling activity, combined with habitat assessments, establishes amphibian population status and site quality. Data collection should follow standardized protocols for sampling effort and replication to maximize comparability and detectability (Strong et al., 2017).

Laboratory assessments characterize amphibian health by measuring physiological, immunological, genetic, and molecular indicators and offer insights into the quality of aquatic and terrestrial environments as well as connectivity between them. Recognition of specific stressors guides the choice of parameters and sampling protocols. For example, elevated temperature and sedimentation bias metabolic activity towards

amphibians unable to complete larval development, leading to early-stage mortality. Consequently, metabolic indicators—such as glucose, lactate, and total protein—are routinely analyzed in studies investigating links between these stressors and amphibian health.

A variety of robust amphibian biomarkers characterize physiological and immunological condition. Elevated concentrations of stress hormones—particularly glucocorticoids—indicate disruption of natural-secretory patterns by environmental perturbation. Additionally, immune markers such as total white blood cell counts serve as indicators of immune activation, while assessment of immune-cell subtypes can reveal amplification or suppression of specific immune functions. Other physiological or metabolic indicators related to water quality (e.g., pH, dissolved oxygen, salinity, nutrients) and habitat quality (e.g., toxicity, contaminants) prove valuable for monitoring and routine water quality assessment. All of these indicators are measured as relative changes compared to control site or predetermined baseline data.

Molecular genetics and pathogen evaluation techniques identify and quantify the presence of several contaminants at various environmental and biological temperatures. Three general domains of molecular analysis—gene expression, genotyping, and pathogen detection—enhance insights into amphibian population dynamics and species diversity. Specific uptake ratios denote the exposure of amphibians under different feed ration levels. Quality control for these molecular data types includes selection of a controlled experimental design, subgroup or comparative analyses at the data-acquisition stage, and standardized basic human-society values.

4.2.1. Physiological and Immunological Biomarkers

Stress hormones, immune markers, and metabolic indicators represent chemical signals emitted by amphibians under environmental strain, each providing a different perspective on the potential sources and consequences of stress (Ceccato et al., 2016). In determining the health status of an individual amphibian in a specific context, biomarker concentrations are placed in context with the stressor by considering predicted stressor ranges and the established biological roles of each chemical. For example, elevated stress hormone concentrations are diagnostic of exposure to limited food resources or excessive environmental disturbance, while low concentrations accompanied by a significant parasite load indicate a broken immune system.

4.2.2. Genetic and Molecular Approaches

Genetic and molecular approaches complement established physiological assessments of amphibian health and habitat quality by enhancing sensitivity to pollution and other

environmental perturbations (Strong et al., 2017). These methods provide additional indicators of stress through gene expression profiling, genotyping, and pathogen detection. Extremes of aquatic habitat—permanent ponds or intermittently flooded, heavily degraded agricultural systems—drive river, wetland, and floodplain amphibians toward locally rare, low-hydric-condition generalized-breeding-site microhabitats, retarding recruitment-related tadpole metamorphosis. These select Mendelian loci enable genetic assessment of fine-scale spatial structuration of headwater-linked upland-forest populations subject to perturbations of high-vegetative-cover landscape patches. Linkage analysis through high-significance variable- F_{st} (population differentiation) candidate-gene detection, screening for pathogenic chytrid (*Batrachochytrium dendrobatidis*) occurrence on a nationwide scale, and overall-genomic-inflation screening ensure data quality. Responses of aquatic-leaved macrophytes, sensitive-stage/integrated-tailed tadpoles, and chironomid-density-amplitude-competitive-mature acquiring; hatching-time disruption and suboptimally densified-riparian-vegetative-cover and mesotrophic-interbreeding-impact-only-maintaining-vascular-plant-macro-treatment partitions pest-to-cohabitant preservation through prospective-metric-based-symbiont-semi-simple-filer macro/bacterial switch.

4.3. Data Integration and Indicator Systems

Multi-parameter datasets enable environmental health assessments by combining amphibian population status, physiological condition, habitat quality, and water characteristics. Two statistical approaches facilitate data integration. Generalized linear models fit gauged health components to generate site-level health indices, which, when paired with land-use data, characterize landscape-scale human impacts. Alternatively, multi-indicator health systems allow simultaneous tracking of multiple components without interdependence assumptions. Such systems can deliver overall scores or zoning class indications based on suites of indicator species, providing users with straightforward summary information (Hadi Fikri et al., 2013).

Bio-indicator data from various ecosystems demonstrate the feasibility of gauging multiple anthropogenic pressures through amphibians. Freshwater wetlands link hydrological and water-quality alterations to specific community and physiological responses using multi-marker indices. Forest springs and headwater systems connect nutrient-load shifts and land-use changes to microhabitat-structure adjustments and ordination-analysis community-composition variants. Urban and agricultural gradients assess pollutant mixtures, landscape-fragmentation indicators, and ranavirus-disease

prevalence, enabling the evaluation of pressure combinations across an urban-agricultural spectrum (Strong et al., 2017).

5. Case Studies Across Habitats

The influence of different land-use practices on water quality and the related community structure of amphibians in forest springs in southern Brazil have been studied by using a multi-marker bioindicator approach (Shirk, 2010). Riparian areas are essential to the conservation of aquatic ecosystems and their effective management of these areas is key for the protection of headwater streams and the services they provide. To assess the effect of alterations associated with agricultural land-use on headwater springs in the São Paulo State, Brazilian, an integrative indicator framework, based on three relevant response levels (water-quality, microhabitat, and biota) was applied. Water quality monitoring should be integrated with the gathering of information on site- and landscape-scale habitat variables. When shifts in land-use alter pollutant loading it is expected that headwater streams undergo a change from natural geochemical conditions to the locally prevailing agricultural water quality.

Amphibians are facing unprecedented population declines worldwide, due to habitat loss, pollution, invasive species, land-use change and climate alteration. The influence of contaminants on amphibians is often related to life history, Clutch traits, fertilization behaviour etc. The link between water quality and aquatic amphibian population metrics in urban and agricultural landscapes have been assessed using a multi-marker bioindication framework. Responses include verzamelen Metapopulation models have been used alongside Integrated Modelling Systems to describe the role of amphibians.

5.1. Freshwater Wetlands

Amphibian bio-indicator research has highlighted freshwater wetlands as ecosystems where land-use modifications, such as urban development and agriculture, can rapidly alter hydrology and water quality. These changes can generate distinct environmental signatures that influence amphibian assemblage composition, health condition, and population dynamics. The responses of amphibians and associated environmental factors have been monitored in various wetland types within urban and agricultural landscapes, providing insight into the relative impacts of hydrological alterations and nutrient enrichment on amphibian communities and ecosystem structure.

Frogs and toads (Anura) are the ideal bio-indicator clade for assessing environmental alterations in freshwater wetlands. Their complex life cycle and dependence on both aquatic and terrestrial habitats render them particularly sensitive to changes in the land-

water interface in freshwater environments and make them valuable for detecting habitat-quality alterations (H Jouney, 2018).

Complementary bio-indicators sensitive to hydroperiod alteration and nutrient flux within freshwater wetlands include protophyll amphibians such as salamanders (Caudata) and aquatic invertebrates. These taxa further enhance the capacity of the amphibian bio-indicator system to integrate stressor information across multiple ecological dimensions.

5.2. Forest Springs and Headwaters

Several studies have documented frog occurrence in relation to the stream health of forest headwaters (Strong et al., 2017) and, more generally, forest springs (Elizabeth Schneider, 2008) of southern Quebec. These forest headwaters receive significant nutrient fluxes from anthropogenic activities and discharge these nutrients into larger water bodies. Forest streams contribute to nutrient loading through surface and subsurface processes. Environmental monitoring, community composition analyses, and indicator approaches were applied to assess forest headwaters and nutrient-microhabitat interactions. Seasonal and cyclic nutrients regimes, combined with base-flow concentration patterns, were identified, indicating that streams may aggregate, amplify, and buffer the nutrient load and types introduced under climate variability. These studies support the use of different aquatic and semi-aquatic communities, including amphibians, for bio-monitoring policies of nutrient-related challenges specific to spring-fed headwaters and small streams situated at the interface of landscape disturbances. Similarly, forest spring and headwater systems have been used to support a comparative assessment of conducting chemical monitoring in tandem with bio-monitoring (E. Skidds et al., 2007).

5.3. Urban and Agricultural Landscapes

Urban and agricultural land use alters the chemical composition of the environment, introducing a diverse mixture of contaminants into natural habitats. Agricultural contaminants often consist of pesticides and fertilisers, while urban pollutants may include metals, lubricants, hydrocarbons, solid waste, and excess nutrients, among others (Lukanov et al., 2023). Numerous studies have examined the effects of different contaminants in aquatic and terrestrial ecosystems. However, few have assessed the combined effects of these contaminants along gradients of urbanisation or agricultural intensity. Monitoring urban-rural or agricultural gradients can provide insight into the spatial distribution and potential impacts of numerous contaminants across multiple habitats (Li et al., 2020).

Agricultural and urban land use have a strong influence on remaining aquatic habitats, often resulting in the establishment of a distinctive rural gradient. The introduction of synthetic agricultural chemicals, products of industrial processes, and stagnation of residential inhabitants increases the complexity of pollutant mixtures. Consequently, urban-rural gradients represent a promising standard for assessing pollutant mixtures. The intensity of agricultural operation influences the intensity of pollution, with agronomic practices such as tillage, monoculture, and fertilisation further affecting plant beats and other inhibitors. Pollutant type is therefore expected to vary depending on agricultural purpose, requiring a complementary analysis of habitat structure en route to an appropriate indicator.

6. Limitations and Considerations in Using Amphibians as Bio-indicators

The use of amphibians as bio-indicators of environmental health has certain limitations and considerations that must be taken into account in order to ensure accurate and effective monitoring. Firstly, amphibians are a highly diverse and ubiquitous taxon, comprising over 7,000 species that occupy a broad range of habitats. Though bio-indicator programmes that rely on sets comprised only of a few representative species are a practical necessity, biodiversity is not evenly distributed across taxonomic groups and assemblages. Inclusion of ecologically important taxa, such as amphibians, in bio-indicator sets may be limited because they are subject to large short- or long-term population fluctuations that can lead to the misinterpretation of habitat quality and potential for recovery. Furthermore, key knowledge concerning toxicological stress-response relationships involving amphibians has only recently begun to accumulate, meaning that a comprehensive understanding of the impact of entire pollutant mixtures on amphibians is still lacking (Strong et al., 2017). Such knowledge pleads that amphibians should not be the primary focus of multi-species bio-indicator programmes until more information is available.

Secondly, even when appropriate indicators have been identified, environmental assessments frequently operate on larger spatial or temporal scales than those at which changes in amphibian-habitat associations are typically observed. Landscape-scale monitoring may not detect shifts occurring at smaller scales because ecological pressures, notably contamination, will be comparatively less severe. Selection of appropriate landscape types for bio-monitoring thus becomes a paramount necessity. Habitat losses associated with urbanization are generally most pronounced near urban centres, while others indicative of the land-water interface often become more evident at greater distance from the centre. Accordingly, land-management decisions aimed at

conserving costly urban infrastructure will largely target landscape types exhibiting bio-indicator shifts close to the centre.

7. Conservation and Policy Implications

The conservation and policy implications of amphibian indicator systems have been summarized to facilitate incorporation of findings into conservation strategies, regulatory actions, and land-use policies (M Mushet et al., 2012). Water quality indicators have been coupled with nutrient concentrations to propose nutrient thresholds for safeguarding amphibian health in multiple wetland types. Proposed water-quality criteria also underpin recommendations for urban stormwater retrofitting and wetland creation, aimed at augmenting health in regions subject to urbanization.

In a Minnesota landscape subjected to agricultural and urban development, indicator metrics have been employed to assess regulatory compliance, reliance on natural resources, and prioritize protection efforts. In landscape corridors vulnerable to development, indicators enable assessment of barriers to dispersal, habitat availability, and habitat connectivity. A multi-scale modelling framework that integrates activities across all land-cover types helps focus conservation resources on areas where amphibian populations would benefit most.

8. Conclusion

Amphibians are reliable bio-indicators of ecosystem health, integrating information across habitats and multiple stressors affecting diverse taxa. Their elliptic life cycle and dependence on aquatic environments render them sensitive to hydrological alterations and poor water quality (Strong et al., 2017). Decreased amphibian populations indicate a continuum of stressors linked to pollution, climate change, habitat degradation, invasive species, and infectious agents, signalling habitat degradation and loss. Consequently, amphibians represent excellent focal taxa for monitoring efforts addressing land-water interface stressors, and an extensive pool of candidate indicators exists at organismal, community, landscape, and watershed levels. Indicator selection depends on conservation objectives and the spatiotemporal context of monitoring.

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