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Bioindicators for monitoring spring water quality

Conservation and preservation of water resources are essential for the sustainability of life and maintenance of ecosystems. Among the water resources, water springs are responsible for the formation and renewal of streams, rivers, and lakes. They are important for balancing the hydrological cycle, maintaining biodiversity, and supplying water for human activities. However, their ecosystem function is compromised by human activities, such as the occupation of recharge areas by economic activities; inappropriate land use practices; elimination of native vegetation in preservation areas; undue disposal of waste and increase in impermeable areas. In this context, it is necessary to develop and adapt methods for evaluating the quality of spring water that can measure the existing environmental impacts and propose preventive and corrective measures. It is essential to unite results of traditional analysis methodologies with the use of bioindicators. This research aims to evaluate bioindicators that can be used to assess spring water quality, qualifying a potential tool for environmental analysis and monitoring. This is a qualitative and descriptive research of the integrative literature review type. Google Scholar, Capes Journal, and Scielo databases were consulted; the keywords used in the search were bioindicators and water springs. The results show that benthic macroinvertebrate organisms, Artemia salina, Daphnia magna, vascular plants and bryophytes and Tradescantia pallida can be used as bioindicators for assessing spring water quality, mainly benthic macroinvertebrates. The results of this research provide solid bases for the implementation of national programs for biomonitoring of water resources, combined with traditional analyses of water quality.

Keywords: Water resources; Biomonitoring; Biodiversity; Sustainability.

Bioindicadores para monitoramento da qualidade da água de nascente

A conservação e preservação dos recursos hídricos são fundamentais para a sustentabilidade da vida e manutenção dos ecossistemas. Dentre os recursos hídricos, as nascentes são responsáveis pela formação e renovação de córregos, rios e lagos. São importantes para equilibrar o ciclo hidrológico, manter a biodiversidade e fornecer água para atividades humanas. No entanto, a sua função ecossistêmica é comprometida por atividades antrópicas, tais como a ocupação de áreas de recarga por atividades econômicas; práticas inadequadas de uso da terra; eliminação de vegetação nativa em áreas de preservação; descarte indevido de resíduos e aumento de áreas impermeabilizadas. Neste contexto, é necessário desenvolver e adaptar métodos de avaliação da qualidade das águas de nascentes que possam medir os impactos ambientais existentes e propor medidas preventivas e corretivas. É fundamental unir resultados de metodologias tradicionais de análise com a utilização de bioindicadores. O objetivo da presente pesquisa é verificar quais os bioindicadores que podem ser utilizados para avaliar a qualidade da água de nascentes, qualificando uma ferramenta potencial de análise e monitoramento ambiental. Trata-se de uma pesquisa qualitativa e descritiva do tipo revisão integrativa da literatura. Foram consultadas as bases de dados Google Scholar, Capes Periódicos e Scielo. As palavras chaves utilizadas foram bioindicadores e nascentes de água. Os resultados mostram que os organismos macroinvertebrados bentônicos, Artemia salina, Daphnia magna, plantas vasculares e briófitas e Tradescantia pallida podem ser empregados como bioindicadores para avaliação da qualidade da água de nascentes, com destaque de referência aos macroinvertebrados bentônicos. Os resultados desta pesquisa fornecem bases sólidas para a implementação de um programa nacional de biomonitoramento de recursos hídricos, junto as análises tradicionais da qualidade da água.

Palavras-chave: Recursos hídricos; Biomonitoramento; Biodiversidade; Sustentabilidade.

Topic: **Conservação da Biodiversidade**

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INTRODUCTION

Water resources are essential for the sustainability of life, maintenance of ecosystems, and development of the most diverse human activities, such as domestic use, industrial production, energy generation, agriculture, and livestock (POFF et al., 2003; FAILLA et al., 2021; WEERASOORIYA et al., 2021). These resources are characterized by fresh surface and groundwater available for use. Surface water flows or is stored on the earth's surface, with contribution from precipitations, aquifer recharge, and runoff from surface water bodies, including streams, rivers, and lakes. Groundwater is found in the subsoil and/or soil and is defined as the water that infiltrates and percolates through the soil and rock layers and is stored beneath them, generating reserves called aquifers. Surface water and groundwater are characterized by their instability and mobility in a closed, continuous, and dynamic cycle called hydrological cycle, whose functioning is sequential and depends on different elements, including water sources, responsible for maintaining its balance (ALMEIDA et al., 2021).

Water springs are defined as the natural outcrop of the water table and are responsible for forming and maintaining the perenniality of streams, rivers, and lakes. They are often used as source for supplying human activities. Water sources are important for maintaining the hydrological cycle balance, as they allow the passage of the groundwater stored in the water table to the earth's surface. In addition, the water that flows from springs to the earth's surface is essential for biological and metabolic processes, ensuring the maintenance of biodiversity (ROMERO, 2017; FAILLA et al., 2021).

Despite the ecosystem function of water springs, human activities have been compromising the maintenance of quality and quantity of these waters due to occupation of recharge areas for economic activities; inappropriate land use practices, causing soil erosion; elimination of native vegetation in permanent preservation areas; uncontrolled population growth and an increasing urbanization with poor basic sanitation systems; and inadequate disposal of solid waste and increased impermeable areas (TUCCI, 2008; FAILLA et al., 2021). Damage to water quality caused by human activities is one of the world's major problems and demands the development and adaptation of methods for assessing environmental quality (PRESTES et al., 2019). Environmental monitoring programs are instruments that enable the generation of standard reference situations, which essentially consist of specific measurements and observations to assess the occurrence of certain environmental impacts and measure their intensities, in addition to evaluate the effectiveness of implemented preventive measures (BITAR et al., 1998; FORIO et al., 2020).

Traditional environmental monitoring uses physical, chemical, and microbiological variables for immediate identification of changes in water characteristics, with accuracy in determining altered concentrations. However, it does not show the totality of the environmental conditions of these aquatic resources, but only the state of the water at the time of collection (CALLISTO et al., 2004; FORIO et al., 2020).

Applying integrated analyses of water quality by combining traditional methodologies that analyze physical, chemical, and microbiological variables with biological aspects of the system is essential for efficient evaluations, providing a more sensitive and predictive analysis of environmental impacts. The application of biological variables is based on the responses of organisms to the environment. The aquatic biota reacts to the different disturbances to which they are subjected. Thus, a comprehensive characterization through biological indicators, known as bioindicators, shows the interaction with the physical environment and allows the study of the entire aquatic community and its responses to stress resulting from anthropogenic actions that gradually affect water resources (MA et al., 1994; PARMAR et al., 2016; PRESTES et al., 2019).

The use of bioindicators to assess water quality is an established practice in several countries. The Environmental Protection Agency of the United States (EPA) suggests the use of a specific group of organisms or one organism to characterize the ecological system conditions in an impacted or little impacted area, combined with evaluation of traditional water quality parameters (BARBOUR et al., 1999). Regarding the European Union, the norms for assessing the quality of water resources are based on the use of different bioindicators, such as phytoplankton, macroalgae, angiosperms, benthic invertebrates, and fish. Physical and chemical environmental conditions are evaluated after these analyses. Considering the set of evaluations, biomonitoring is initially carried out by toxicological tests to confirm whether the contamination is a concern for the life of organisms in this ecosystem (ROBERTO, 2014).

According to Callisto and coauthors (2004), changes in nature define the bioindicators to be used in a study, which can distinguish the instability between natural and anthropogenic changes, showing the frequency and proportion of impacts on the ecosystem (CALLISTO et al., 2004). A bioindicator species has a low tolerance to variations in its environment, and its presence in an area indicates the specific conditions of that environment. Thus, changes in the environment cause variations in the species population (PARMAR et al., 2016; FORIO et al., 2020; PRESTES et al., 2019).

Studies have shown that use of bioindicators is efficient to assess environmental quality at low cost, in addition to enabling the detection of effects of toxic agents on the existing biota. Therefore, monitoring contaminants found in other groups of individuals enables the determination of the environmental contamination increase and threat. Biological tests used to investigate the toxicity of environmental contaminants mainly use plants, animals, and human cells as bioindicators (PARMAR et al., 2016; PRESTES et al., 2019).

Biological monitoring is based on changes in the structure and composition of aquatic organism communities. However, the response time of several groups of organisms in the environment can be considerably long (years to decades), thus, specific groups (protozoa, ciliates, algae, benthic macroinvertebrates, and fish) have been selected for the obtaining of immediate responses, using different analysis methods (PARMAR et al., 2016; FORIO et al., 2020).

Traditional water assessment does not reflect the environmental conditions of these aquatic resources, but only the state of the water at the time of sample collection. Combining traditional methodologies with the use of bioindicators is essential for advances in the analysis of spring water quality. Biological assessment involving the integrity of these ecosystems can represent an efficient systemic analysis strategy for developing and planning actions focused on the balance between socioeconomic development and protection of these ecosystems (PARMAR et al., 2016).

This research aims to provide an integrative literature review using the main world databases to analyze potential bioindicators of spring water quality, in addition to subsidize government agencies in the implementation of a national program for biomonitoring of water quality combined with traditional analysis methods, providing a more comprehensive tool for analysis and monitoring of water resources.

MATERIALS AND METHODS

This study consists of a qualitative, descriptive, integrative literature review for search, critical evaluation, and synthesis of evidence on the investigated topic. The research was composed of the following stages: choice of theme and construction of the research question; sampling; data collect; critical evaluation and categorization of selected studies; interpretation of results; and synthesis of the information obtained (SOUZA et al., 2010).

Exclusion and inclusion criteria for sampling were defined, in addition to the descriptors to be used for searching the databases listed for the selection of articles. Inclusion criteria were complete original articles available for free online, published from 2015 to October 2020, in Portuguese, Spanish, or English, corresponding to the research objective. Exclusion criteria were articles unrelated to the proposed theme, duplicated, incomplete, published in languages other than Portuguese, English, or Spanish, and not available fully online. The following material were also excluded from the study: theses, dissertations, review articles, pilot studies, letters, editorials, and communication. An active search in Google Scholar, Capes Journal, and Scielo databases was based on the following keywords, in the three languages: bioindicators, water, environment, and springs.

The selected articles were fully read and then analyzed through a critical evaluation based on categorization, using the following variables: bioindicators used, author and year of publication, objective of the study, conclusions of the research, and the environmental conditions in which the bioindicator was evaluated. Finally, the results were interpreted by developing a more detailed study on the bioindicator organisms identified in this research.

RESULTS

A total of 2987 articles on the subject were found in the searched databases: 109 in Scielo, 2320 in Google Scholar, and 558 in Capes Journal. Two records were excluded due to duplication. The title and abstract of the articles were read and those that met the inclusion criteria and answered the guiding question were selected (Figure 1).

The use of the keywords bioindicators, water, environment, and springs as the first search filter showed the need for delimiting the research field, as water and environment encompasses a wide range of studies (air, oceans, rivers, volcanoes). Thus, a second filter was applied to the search, using the keywords bioindicators and springs; then, 1487 articles were found in Google Scholar and Capes Journal databases. Regarding Scielo database, a third filter was needed, using the keyword springs alone to access more qualified texts, which enabled the obtaining of 106 articles. Therefore, 1593 articles were found in the three databases.

Subsequently, the articles were selected based on information in the title and abstract that effectively addressed the study topic and answered the guiding question: what are the bioindicators used to assess the spring water quality? In this sense, 45 articles were selected for full reading, and eight of them provided quality answers to the guiding question.

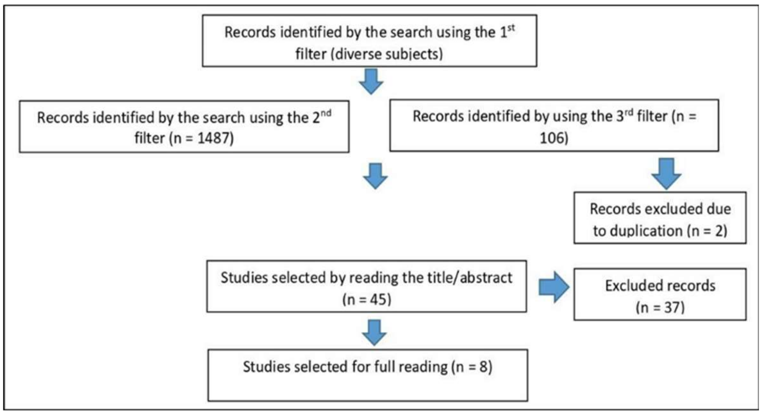

Figure 1: Flowchart of the processes of research and selection of articles for review.

The classification of the selected articles according to year of publication, research location, and bioindicator used to assess the quality of water for human consumption are summarized in Table 1.

Regarding the methodological approach, all selected studies are quantitative. Regarding to material and methods, all the eight selected studies (100%) were developed exclusively in springs. Regarding the location of the studies for assessment of environmental quality of springs using bioindicators, six studies (75%) evaluated springs in Brazil and two (25%) evaluated springs in Europe (Italy and Poland).

Considering the studies selected to identify potential bioindicators, 2 (25%) used macroinvertebrates (SANTOS et al., 2017; THAPA et al., 2020), 1 (12.5%) used chironomids (LENCIONI et al., 2018), 1 (12.5%) used vascular plants and bryophytes (PUCZKO et al., 2018), 1 (12.5%) used Tradescantia pallida var. purpurea Boom (RUBIO et al., 2015), 1 (12.5%) used Artemia salina (CEMBRANEL et al., 2019), 1 (12.5%) used Oligofauna (LOPES et al., 2019) and 1 (12.5%) used Daphnia magna (MOURA et al., 2018).

The analysis of the eight selected articles contributed to the identification of animals and plants that are used as bioindicators to monitor spring water quality through biological evaluation. Thus, the analysis and discussion of bioindicators are pertinent for their use in environmental monitoring programs.

Table 1: Summary of the articles selected for full reading and identification of the bioindicators used.

Bioindicators	Title	Author	Objective	Conclusion
Benthic macroinvertebrates	The influence of land use and occupation on spring water quality - analysis of benthic macro invertebrates as bioindicators.	SANTOS et al. (2017)	Analyze the influence of land use and occupation on the water quality of the Machado de Melo stream sub-basin in the municipality of Araçatuba/SP, using benthic macroinvertebrates as bioindicators.	The human pressure on the biota of the Machado de Melo stream sub-basin reflects the impact of the benthic macroinvertebrate community, mainly with a predominance of organisms from resistant and/or tolerant groups, considered indicators of pollution.
Benthic macroinvertebrates	Assessment of spring water quality in Jhimruk River watershed, lesser Himalaya, Nepal.	THAPA et al. (2020)	This study assesses the spring sources to clarify concerns about the quality and safety of this water used for human use, as well as the condition of the biological habitat in the Jhimruk River Watershed (JRW), Nepal.	Ecologically, diverse sets of macroinvertebrates were recorded from our spring ecosystems and these animals were signifcantly infuenced by discharge rate, elevation, EC, and nitrate concentrations. We believe that drying and degrading water quality are one of the biggest threats for all dependent on the spring water

				resource. Consequently, there is needed for proper plan to conserve spring sources so that their biodiversity, as well as the water quality of springs, will be adequately managed for sustained use.
Vascular plants and bryophytes	Vascular plant and bryophyte species richness in response to water quality in lowland spring niches with different anthropogenic impacts.	PUCZKO et al. (2018)	Compare the vegetation of niches of floodplain springs in areas with different degrees of anthropogenic transformation, as well as to study the impact of water quality on phytocenosis of springs.	They show that anthropogenic activity significantly affects the biodiversity of plant communities in floodplain headwaters. The presence or absence of crenophytes and bryophytes is indicative of the ecological status of the underground effluent complexes.
Chironomids	Effects of human impacts on diversity and distribution of chironomids (Diptera: Chironomidae) in prealpine springs.	LENCIONI et al. (2018)	Analyze the distribution and diversity of chironomid fauna in thirty-six pre-alpine springs in relation to altitude and their conservation status from untouched to highly disturbed.	The usefulness of chironomids as bioindicators of water quality and the ecological status of springs was confirmed, with some species associated with high disturbance for water intake works and others with primitive conditions.
Tradescantia pallida var. *purpurea* Boom	Integrated Environmental Assessment of streams in the Sinos River basin in the state of Rio Grande do Sul, Brazil.	RUBIO et al. (2015)	It represents the first initiative of an integrated assessment of the environment along the springs of Estancia Velha / Portao, Riachos Pampa and Schmidt using physical, chemical, and biological parameters. The data can contribute to an integrated environmental diagnosis of the Rio dos Sinos basin, supporting public management and providing bases and methods for environmental control.	The results of the application of the rapid assessment protocol (PAR) indicated a natural environmental situation, in terms of habitat diversity and environmental impacts, in the three sampled sites. The bioindicator Tradescantia pallida (Rose) D.R. Hunt var. purpurea Boom showed water genotoxicity in two sampled sites.
Artemia salina - Crustaceans	Environmental quality of spring in urban area	CEMBRANEL et al. (2019)	Evaluate the water quality of a spring located in the urban perimeter of the city of Francisco Beltrao-PR, through physical-chemical, microbiological, and toxicological analyses, in addition to macroscopic environmental analysis of the spring.	The toxicity analysis using A. salina indicated some toxicity in the spring water. The Environmental Impact Index showed terrible conditions for the preservation of the spring.
Oligofauna	Water quality in springs in the city of Araraquara-SP: an approach using environmental bioindicators.	LOPES et al. (2019)	Evaluate the water quality in two springs (points in Corrego do Tanquinho and Corrego da Caixa D'agua) in the urban perimeter of the municipality of Araraquara-SP through the study of oligofauna.	The spring called Corrego da Caixa D'agua (NCD) indicated a precise degree of impact, showing a punctual process of environment degradation. On the other hand, the headwaters Corrego do Tanquinho (NT) did not present a significant stage of pollution.
Daphnia magna	Water quality assessment of springs in the Andreas Basin, RS, Brazil, based on ecotoxicological and genotoxicological assays with *Daphnia magna*. (Straus, 1820)	MOURA et al. (2018)	Evaluate the water quality of springs in the Andreas Basin, RS, using the conventional approach, as well as ecotoxicological and genotoxicological assays, using the Comet Assay (EC) with the test organism *Daphnia magna*.	The results showed differences between the genotoxicological tests and the conventional tests, since by the genotoxicological analyzes, 47.4% of the sampled points were considered toxic, while by the conventional analyses, all these points were classified as classes 1 or 2 by CONAMA (National Environment Council), considered to be of good quality.

DISCUSSION

The bioindicators can be classified into pointing organisms, test organisms or monitor organisms. Pointer organisms, also known as ecological indicators, show the pollution impact through changes in population size or through existence or disappearance of the organism. Test organisms, are those established and applied in laboratory tests. Monitoring organisms show the pollution impact on living organisms, providing qualitative and quantitative data (KLUMPP et al., 2001; BRAZ, 2020).

According to the analysis of the present study, benthic macroinvertebrates were the most used bioindicators of water quality. Two studies used benthic macroinvertebrates (BM) to assess environmental impacts on water sources. Santos et al. (2017) carried out a qualitative and quantitative study on BM fauna by evaluating the density of organisms in each sample. The researchers concluded that the analysis of the richness and abundance of species in the water and in the sediments allows an interconnection between the areas with more intense environmental impacts, together with a reduction in the protective riparian forest with the abundance of tolerant organisms (indicators pollution) and the absence of sensitive organisms. Thapa et al. (2020) evaluated Macroinvertebrat communities data using the Vegan package in R studio, Spearman Rank Correlation between Bray-Curtis similarity of species data and the abiotic data matrix (function bioenv in R), Canonical Correspondence Analysis (CCA). Also there used the Spearman correlation analysis to compare the WQI with selected macroinvertebrate metrics including family richness, Ephemeroptera+Plecoptera+Trichoptera (EPT) richness, % EPT individuals, and % Chironomidae individuals (Table 1) (THAPA et al., 2020). The Ephemeroptera+Plecoptera+Trichoptera (EPT) richness, % EPT individuals, and % Chironomidae individuals are biological metrics to assess an environment quality (SANTOS et al., 2016). The researchers concluded that the macroinvertebrate assemblages' structure was infuenced most by discharge rate, elevation, EC, and nitrate concentration (THAPA et al., 2020).

Benthic macroinvertebrates stand out among the biological variables used to assess environmental impacts, from different anthropogenic actions, on water quality in different aquatic environments (PIMENTA et al., 2016; HAGEMEYER et al., 2022). They are invertebrates that can be retained in a 0.2-mm mesh, represented by several Phyla, such as Arthropoda (insects, mites, crustaceans) Mollusca (gastropods and bivalves), Annelida (oligochaetes and leeches), Nematoda, and Platyhelminthes. Insects stand out due to their diversity and abundance, whose distribution depends on morphometric, physical, and chemical characteristics of the habitat and food availability. Arthropoda, Mollusca, Annelida, and Platyhelminthes species are usually associated with bottom habitats of surface beds and lakes, whether they are fixed to a substrate, called benthic community (PIMENTA et al., 2016).

The wide use of benthic macroinvertebrates as bioindicators of water quality is justified by their characteristics: long life cycle, as they can live for weeks, months, and even more than one year; they are usually large (greater than 125 or 250 µm), sessile or poorly mobile organisms, which facilitates sampling, thus requiring a relatively low-cost evaluation procedure; great taxonomic diversity and relatively easy identification (at level of family and some genera); and are organisms sensitive to different pollutant concentrations in the environment (CALLISTO et al., 2004; FORIO et al., 2020; HAGEMEYER et al., 2022).

Benthic macroinvertebrates are variably sensitive to environmental conditions, sedentary, and able to absorb the environmental conditions, offering a reliable diagnosis when considering impacts on organisms (CALLISTO et al., 2004). They are classified as resistant, tolerant, or sensitive to pollution. Resistant benthic groups, such as species of Tubificidae and chironomid larvae, are insensitive to a several environmental stresses and can reach high population densities in polluted rivers. Tolerant species, such as Odonata larvae and adult Coleoptera and Hemiptera, are also able to survive in a wide range of environmental conditions, but they do not survive in environments under severe stress. Sensitive or intolerant benthic organisms, such as Plecoptera, Ephemeroptera, and Trichoptera larvae, grow and develop under narrow environmental conditions and are rarely found in polluted rivers. The presence of Plecoptera, Ephemeroptera, and Trichoptera indicates clean water; thus, they have been used in the northern hemisphere as bioindicators of water quality (FRANÇA et al., 2019; PRESTES et al., 2019; FORIO et al., 2020).

Benthic macroinvertebrate communities are also used to evaluate effects of copper and zinc metals, as these metal ions are in fertilizers and pesticides and in organic waste from urban areas (PIMENTA et al., 2016). In this sense, they can be used for identifying impacts and analyzing water quality in urban and rural drainage. A similar study was developed by Colpo et al. (2009), who used benthic macroinvertebrates as indicators of environmental impacts caused by effluents from rice crop areas and urban and industrial activities; they found that effluents released by urban and industrial activities are more aggressive to aquatic environments than those generated by rice farming (COLPO et al., 2009).

The evaluation of changes in water quality in environments under different anthropogenic actions (rural and urban areas) using benthic organisms as bioindicators combined with analysis of water and sediments provides more robust and safer diagnoses and, consequently, allows for better interpretation of the results (PIMENTA et al., 2016).

CONCLUSIONS

This research provides an integrative literature review of the literature available in the main world databases to asses potential bioindicators of spring water quality.

The results show that benthic macroinvertebrate organisms, *Artemia salina, Daphnia magna*, vascular plants and bryophytes and *Tradescantia pallida* can be used as bioindicators of spring water quality, mainly benthic macroinvertebrates. However, each organism has specificities as a potential bioindicator of spring water quality.

Many countries stablished the use of bioindicators as an environmental monitoring instrument for assessing water quality. The results of this research provide solid bases for the implementation of national programs for biomonitoring of water resources, combined with traditional analyses of water quality.

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