



Exploring fungal potential for enhancement of environment

Explorando o potencial dos fungos para a melhoria do meio ambiente

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ABSTRACT

Inadequate effluent disposal has caused damage to the environment worldwide. This study aimed to perform a scientometric analysis of studies exploiting fungi applied to improve the quality of effluents. We used *Web of Science*, *Scopus* and *Pubmed* databases to search for publications between 1980 and 2023. The pollutants and effluent quality parameters most commonly addressed in scientific literature were identified, revealing trends and gaps in the field. A correlation analysis was performed between the variables Research and Development (R&D), gross domestic product (GDP) per capita, and number of inhabitants per country to investigate whether these variables are correlated with the number of research studies in each country. In addition, a linear regression was performed to investigate the effect of the number of inhabitants per country of each country on the number of studies. A total of 11183 articles were obtained, of which 2001 were identified as related to the main topic, and then more than 30 different types of pollutants were removed, such as primarily including dyes (951), phenolic compounds (682), and heavy metals (562). Concerning effluent quality parameters, chemical oxygen demand was most frequently mentioned in the articles obtained from the literature review (620). The world's most populous countries produce the largest number of studies related to the topic. Our results highlight the bioremediation potential importance of fungi in the scientific literature, even under inhospitable conditions for microorganisms (such as toxicity, low temperatures, and high acidity), reducing environmental damage in aquatic environments and mitigating harm to public health.

Keywords: water; bioremediation; pollutant; effluent; scientometrics.

RESUMO

O descarte inadequado de efluentes tem causado danos ao meio ambiente em todo o mundo. O objetivo deste trabalho foi realizar uma análise cienciométrica de estudos que buscaram melhorar a qualidade de efluentes com a aplicação de fungos. As bases de dados *Web of Science*, *Scopus* e *PubMed* foram utilizadas na busca dos periódicos, publicados entre 1980 e 2023. Os poluentes e parâmetros de qualidade dos efluentes mais abordados pela literatura científica foram identificados, apontando tendências e lacunas da ciência. Foi realizada uma análise de correlação entre as variáveis: desenvolvimento & pesquisa (P&D), produto interno bruto (PIB) *per capita* e número de habitantes por países para investigar se essas variáveis estão correlacionadas com o número de pesquisas em cada país. Além disso, este estudo investigou o efeito do número de habitantes de cada país sobre o número de estudos por meio de uma regressão linear. Foram encontrados 11,183 artigos e, após a leitura de resumos, 2,001 abordavam o tema. Mais de 30 tipos diferentes de poluentes foram removidos, principalmente corantes (951), compostos fenólicos (682) e metais pesados (562). A respeito dos parâmetros de qualidade do efluente, a demanda química de oxigênio (DQO) é o que aparece na maior quantidade de artigos (620). Os países mais populosos do mundo produzem maiores quantidades de estudos relacionados ao tema. Este trabalho demonstrou o potencial biorremediativo dos fungos na literatura científica, até mesmo em condições mais inóspitas a micro-organismos (toxicidade, baixas temperaturas, acidez elevada), reduzindo danos ambientais em ambientes aquáticos e prejudiciais à saúde pública.

Palavras-chave: água; biorremediação; poluente; efluente; ciencimetria.

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Introduction

Global water consumption has nearly doubled in the last few decades; meanwhile, increasing concerns about water contamination have led to extensive research and development of water treatment techniques (Latif et al., 2023). These techniques are expanding to promote water reuse and improve water quality for human consumption. However, water pollution is a global problem that poses a serious threat to the survival of all forms of life. In addition, aquatic pollution can be caused by organic and inorganic impurities, as well as microbiological contaminants (Sharma, 2023b; Yusuf et al., 2023). Likewise, water pollution is caused by various anthropogenic factors such as population growth, industrial and mining activities, sewage and wastewater, radioactive waste, chemical fertilizers, pesticides, and urban development. Therefore, wastewater discharged into the environment is harmful to human health, animals, and plants (Selim et al., 2021; Umar et al., 2023).

Traditional wastewater treatment methods, such as the activated sludge process, coagulation, flocculation, and adsorption, have been used to solve problems related to wastewater pollution (Zhang et al., 2021). Although conventional methods have proven effective in treating common environmental pollutants, they are not effective in the removal of complex pollutants and the production of environmentally toxic by-products (Latif et al., 2023). In addition to this bottleneck, conventional methods produce a variety of secondary pollutants during the degradation process that are harmful to the environment (Selim et al., 2021; Zamri et al., 2021; Latif et al., 2023).

As a result, novel and innovative treatment methods have been explored. Many of these newly developed techniques involve biological methods using microorganisms (fungi, algae, and bacteria), due to their natural ability to biodegrade and degrade complex pollutants. These methods are less expensive and less harmful to the environment (Sundararaju et al., 2020; Kumar and Dwivedi, 2021; Latif et al., 2023). Wang (2023) emphasizes that fungi offer more advantages than other microorganisms in the removal of pollutants from wastewater. In addition, they secrete numerous extracellular enzymes such as cellulase, xylanase, pectinase, laccase, peroxidases, and many others. Also, fungi are a rich source of bioactive metabolites that have significant biotechnological potential (Chatterjee et al., 2020; Negi and Das, 2023).

Fungal wastewater treatment systems are an excellent solution to remove contaminants from water, especially after the emergence of new recalcitrant and complex pollutants that cannot be treated by conventional technologies (Sundararaju et al., 2020). Likewise, they are resistant to difficult adverse conditions such as high suspended solids loads, extreme pH variations, low tolerance to nutrient concentrations, and the presence of organic pollutants and heavy metals (Espinosa-Ortiz et al., 2016). Furthermore, fungi contribute to the production of a wide range of different metabolites, such as citric acid, lovastatin, lactic

acid, carotene, extracellular polymers, and enzymes, which make them more attractive for bioremediation processes (Sharma et al., 2023a).

Therefore, this study focused on studies using fungi to remove pollutants from wastewater from industry, various municipal activities, manure, and agriculture, solid or liquid culture media, aqueous solutions, and synthetic wastewater. The distribution of these studies worldwide and the factors influencing their development were analyzed. In addition, we present benchmark data for mycoremediation research.

Materials and Methods

Data collection

All publications that applied fungi, community of microorganisms present in soil, sludge and mushroom compounds were considered as treatment. Subsequently, for this purpose, the following questions were addressed: Which pollutants were investigated? What quality parameters were monitored? What is the geographical distribution of the published studies? What factors influence the number of publications by country? Initially, we conducted a literature survey accessing different scientific databases such as Web of Science, Scopus and Pubmed. Then, we use information provided at title, abstract, and the following keywords: ((fung* OR mushroom*) AND (remediation* OR bioremediation* OR mycoremediation* OR treatment* OR remov* OR biosor*)) (wastewater* OR sewage* OR effluent*). Finally, filters were applied for only “articles,” to find published papers covering the period from 1980 and 2023. The data were obtained through pre-selection based on the abstracts.

Studies by country

Data from effluent recording points were obtained from the literature survey, and represented on maps, delineated by countries, using the software QGIS v.3.22.8 (QGIS Development Team, 2023). The articles were classified according to pollutants and quality parameters, and the results were plotted using graphics. The X and Y-axis scales were adjusted to improve the visual representation of the data.

Statistical analysis

A correlation analysis and linear regression between the variables Research & Development (R&D), GDP per capita, and the number of inhabitants by countries were performed to investigate whether these variables are correlated with the number of research studies in each country. The GDP per capita was evaluated to investigate the influence of the level of economic wealth by country on the number of articles. The correlation was represented through principal component analysis (PCA), and a collinearity assessment was conducted to determine the regression model.

Initially, the degree of collinearity among the explanatory variables was examined by estimating the Variance Inflation Factor (VIF), with values calculated using the “USDMM” package (Naimi, 2014).

Consequently, all variables were retained in the analysis as they exhibited values ≤ 10 , as recommended by O'Brien (2007). This was complemented by a Durbin-Watson (DW) autocorrelation analysis, with values ranging from 0 to 4; values equal to 2 indicate zero autocorrelation; those less than 2 indicate positive autocorrelation, and more than 2 indicate negative autocorrelation, as established in the "Car" package (Fox and Weisberg, 2019).

To assess the list of explanatory variables established above concerning the number of articles by country, a generalized linear model was employed with a Poisson error distribution. Additionally, an ANOVA with the chi-square test was conducted to assess the statistical significance of the models. The model reduction was achieved through permutation tests (1,000 permutations), and its explanatory power was evaluated based on the adjusted determination coefficient (R^2 , R^2 - adj) proposed by Peres-Neto et al. (2006), and implemented in the "vegan" package. All analyses were carried out using the R language v.4.3.2 (R Core Team, 2023).

Subsequently, a simple linear regression was performed between the variables that were deemed representative and best explained the variation in the number of research studies per country. All analyses were conducted using the R language, version 4.3.2. (R Core Team, 2023), Research & Development, GDP per capita, and population size data were obtained from The World Bank platform, available at: <https://www.worldbank.org/en/home>.

Results and Discussion

Contaminants, pollutants, and wastewater quality

We found 11,183 articles related to mycoremediation research, of which 2,001 revealed 35 classes of pollutants downstream of the WWTP, which were tested for removal by fungi. Most studies investigated the removal of dyes (951), followed by phenolic compounds (682), heavy metals (563), and pharmaceutical compounds (400) (Figure 1). Dyes, phenols, and heavy metals are the main targets of bioremediation research due to numerous sources of pollution such as textile, paper, plastic, printing, leather, and paper industries (Bharathi et al., 2020). Moreover, these pollutants exhibit high durability, stability, and resistance to degradation. Also, they are highly toxic and carcinogenic, and can cause cellular mutations (Jun et al., 2019), while heavy metals bioaccumulate in living organisms (Genchi et al., 2020). The concern of the community and public authorities for public and environmental health drives scientific research.

Ten quality parameters downstream of the WWTP were identified in the analyzed literature. COD (chemical oxygen demand) was the most frequently reported (620), followed by effluent color (55), biological oxygen demand (BOD) (29), total organic carbon (TOC) (20), and total suspended solids (TSS) (15) (Figure 2). Similarly, COD is the topic of greater interest in scientific research, reflecting the problem of population growth that generates effluents with high organic matter, which can come from the industrial, agroindustrial, and domestic sectors.

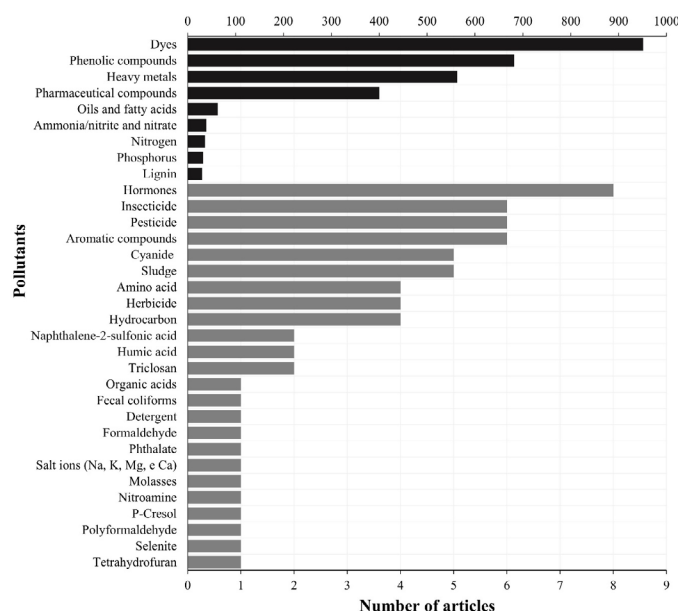


Figure 1 – Pollutants removed by fungi in culture media (solids and liquids), aqueous solutions, artificial effluents, as well as effluents from industries, agriculture, shocks, and domestic, in the scientific literature. The black upper axis represents the number of articles between 10 and 1000, while the gray lower axis represents the number of articles between 0 and 9.

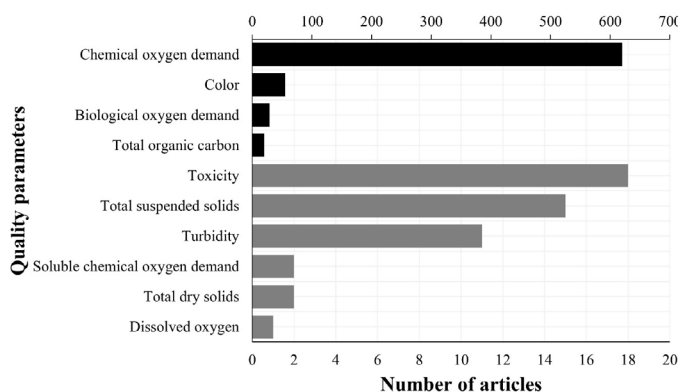


Figure 2 – Improvement of quality parameters by fungi in culture media (both solids and liquids), aqueous solutions, artificial effluents, as well as effluents from various sources, including industries, agriculture, slurry, and domestic wastewater, as documented in the scientific literature. The black upper axis represents the number of articles between 10 and 700, while the gray lower axis represents the number of articles between 0 and 20.

COD is a measure of the amount of oxygen, divided by the volume of the system, required to oxidize organic (and inorganic) matter in wastewater using a chemically oxidizing agent, expressed in milligrams of O₂ per liter (Nagel et al., 1992). It is a quality parameter widely applied in studies that evaluate effluent quality for the following reasons: first, it serves as an indicator of the compliance of wastewater discharge and treatment procedures with current standards.

Second, in WWTPs, the ratio of BOD to COD indicates the biodegradable fraction of an effluent. Third, the BOD/COD ratio serves as an indicator of the size of a wastewater treatment plant required for a specific location. Finally, COD only requires a few hours (2-4 h) to measure, while BOD takes five days, making the former more popular and useful than the latter as a water quality parameter (Jouanneau et al., 2014; Li et al., 2018).

In more recent research, two new parameters have emerged: soluble Chemical Oxygen Demand (COD) (Islam et al., 2019) and mineral salts (Na, K, Mg, and Ca) (Ellouze et al., 2010; Sharma and Malaviya, 2016; Gao et al., 2020). Sharma and Malaviya (2016), who used *Cladosporium perangustum* and *Penicillium commune* fungi to remove sodium (Na^+) from leather industry effluents, explain that this chemical element is crucial for the growth of fungal biomass. Another pollutant investigated only in recent research is the herbicide Dichlorofenoxiacetic acid (2,4-D), which was removed from an aqueous solution by *Pleurotus ostreatus* at 99.6% efficiency (Pereira et al., 2023). Four other studies were found that applied fungi to remove herbicides (Loffredo et al., 2016; Marinho et al., 2017; Zeng et al., 2017). Pesticides are also featured only in recent studies (Levio-Raiman et al., 2021; Rajpal et al., 2023). In 2023, at the Web of Science database, the first study that removed fecal coliforms (Wang et al., 2023) was recorded, which used mycorrhizal fungi from the Canna plant. It is possible that new pollutants will be investigated over time, following the development and evolution of human activities.

In the study by Gao et al. (2020), a larger-scale treatment (mesocosms) removed mineral salts (Na, K, Mg, and Ca) and reduced COD at low temperatures (17°C) by treatments with the mycorrhizal fungus *Glomus etunicatum* accompanied by an ecological floating bed. Pollutant removal was not affected by salinity levels. The authors justified that mycorrhizal fungi absorb inorganic elements and degrade organic matter, which enhances plant growth and their tolerance to ion and salt stress, increasing the function of the floating ecological bed when temperatures decline. Only two studies conducted fungal treatment at low temperatures, recommended for Nordic countries (Gonzalez-Martínez et al., 2018; Gao et al., 2020). In a study by Zhou et al. (2023), a reduction in COD in wastewater from oil fields with high salinity was observed using a fungal consortium. More studies are expected to investigate mycoremediation in inhospitable conditions for microorganisms.

Geographic distribution

A total of 549 articles were found on effluents from industrial, domestic, and slurry sources, but 123 publications did not report the origin of the effluent. The incubation of fungi in real effluents illustrates a more viable scenario for their application since in these studies variables such as pH, toxicity, and the presence of other competitive microorganisms are not controlled. The data indicate the world's most populous countries, such as China and India, invest more in the search for alternatives for effluent treatment. In China, 35 studies were found on wastewater bioremediation; in Spain, 30; and in India, 42 (Figure 3).

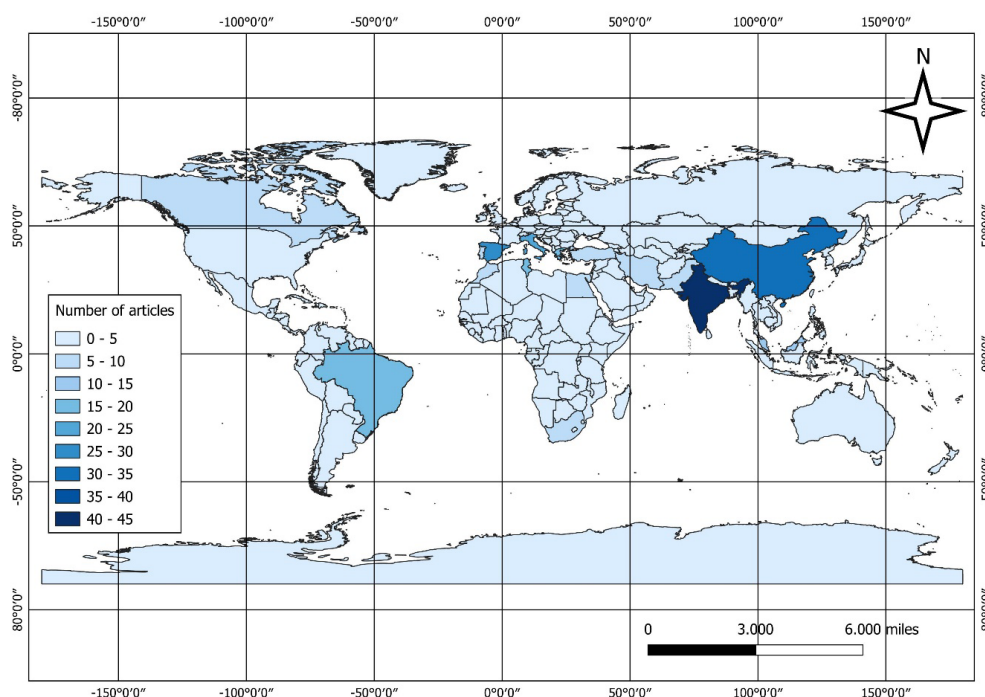


Figure 3 – Representation of countries in which effluents from industries, municipalities, agriculture, and shocks were collected and underwent treatment by fungal cells in the scientific literature.

According to data from The World Bank (2022), China has 1,412,175.00 inhabitants, and India has 1,417,173.17, respectively.

Government actions may have contributed to investments in the conservation and sustainable use of water resources in China, Spain, and India. According to The World Bank, China invested 2.43% of its Gross Domestic Product (GDP) in 2021. Other prominent countries are the United States, with 3.46% of the GDP, and Belgium, with 3.46% of the GDP (The World Bank, 2021a). In contrast, India invested only 0.65% of its GDP in R&D, and Spain invested 1.43% in 2020 (The World Bank, 2021b).

Despite scarce investment in R&D, there have been some governmental actions to improve water quality in India, mainly focused on the Ganges River since 1985. In addition, the National Mission for Ganges Cleanliness was instituted in 2014. The actions focus on wastewater treatment, riparian forest restoration, and the promotion of biodiversity (Cao and Vazhayil, 2022). In the case of Spain, the country is integrated into the European model of water resources management known as the Water Board (Padovesi-Fonseca and Faria, 2022), and receives foreign capital investments.

Unexpectedly, the data in Figure 3 also shows that countries with a high GDP per capita had the lowest number of published papers involving bioremediation by fungi. Li and Zhu (2021) and Furtado et al. (2023) reported that the countries with the highest number of articles on the application of microalgae are China, the USA, and India, according to the scientific literature. García-Ávila et al. (2023) found that the countries with the highest number of publications related to the use of ornamental plants are Brazil, the USA, China, and India.

Authors such as Niknejad et al. (2023) mentioned that the countries with the highest number of studies on green technologies are China, India, Malaysia, the USA, and Australia. However, this study found that the USA and Australia published only three articles on the use of fungi to remove pollutants from wastewater.

Furthermore, a correlation was observed between a country's GDP per capita and the number of articles published on fungal wastewater treatment, as countries with higher GDP per capita tend to publish fewer studies (Figure 4). For example, Australia, Israel, the United States, Canada, Japan, the Netherlands, and France published less than 9 studies. This reinforces the idea that these countries do not focus on the fungal approach. This suggests that these countries have focused more on other biotechnologies, such as microalgae and wetland applications.

Population increase has an impact on the number of articles per country ($p=0.001209$; $R^2=0.73309$; $R^2_{\text{adjusted}}=0.72794$) (Figure 5), and these two variables are strongly correlated (Figure 4). These data suggest that research is being carried out to find solutions to reduce the pollution of water resources caused by population growth, which is linked to industrial expansion, increased energy demand, and worsening problems related to water scarcity. Other factors, such as the economy and public management, can influence the direction of scientific research. However, the sources of pollution generated by the development and growth of the population require public actions to invest more efforts in solving water resource problems.

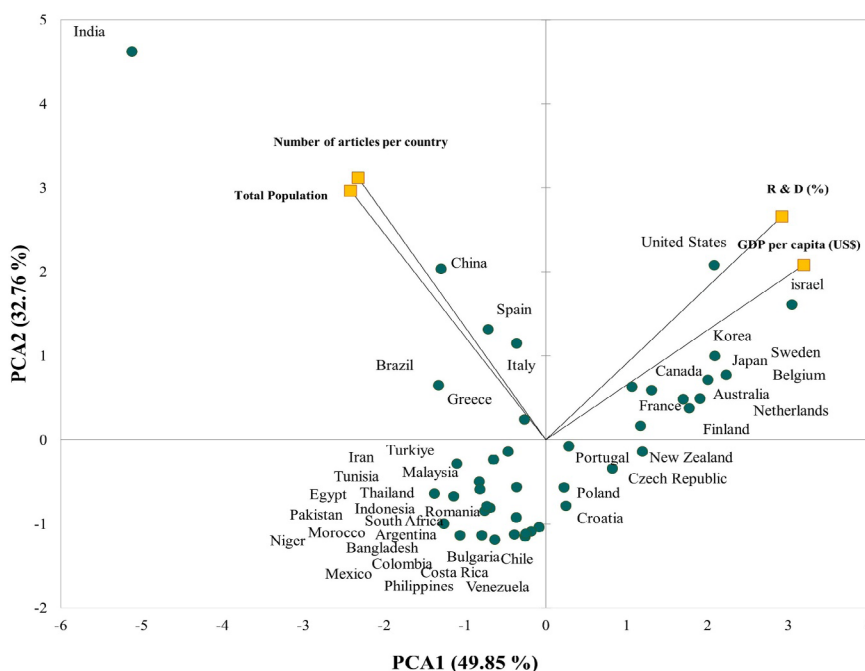


Figure 4 – Evaluation of the influence of variables (R&D, per capita GDP, or population density) on the publication of articles in each country through Principal Component Analysis.

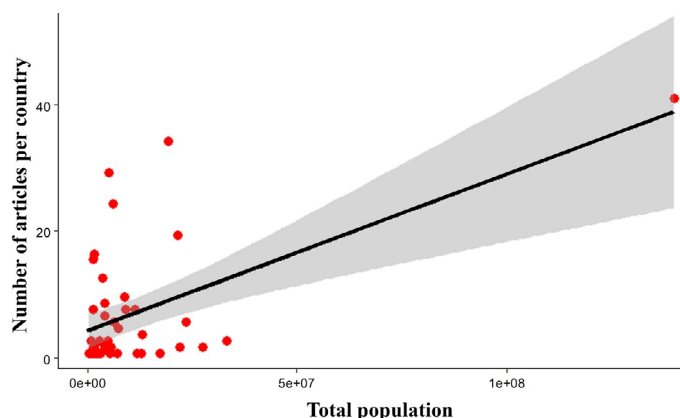


Figure 5 – Effect on the total inhabitants per country in 2022 on the number of published studies regarding wastewater bioremediation by fungi, using linear regression.

Conclusion

Dyes, phenolic compounds, and heavy metals are the focus of scientific research on fungal effluent remediation. Research has demonstrated that it is possible to remove more than 30 different types of pollutants from industrial, domestic, and slurry effluents. A total of 10 quality parameters were identified in the studies analyzed.

Of the 2001 studies, 951 are on colorant removal, 682 on phenolic compounds, and 562 on heavy metals. In terms of effluent quality parameters, chemical oxygen demand is the most frequently mentioned parameter in 620 studies. The reduction of soluble COD, and the removal of minerals, and sodium were research targets only in recent studies, along with fungal resistance to low temperatures, indicating a search for bioremediation solutions in temperate countries. The removal of pollutants at different salinity levels is also a new pre-occupation. The statistical analysis indicates that the number of articles by country is explained by GDP per capita and total population. Linear regression has evidenced that the most populated countries in the world seek bioremediation solutions to improve the quality of their effluent treatment and produce more studies related to the theme. The fungi presented in the potential studies ensure water safety, even in the most inhospitable and difficult environmental conditions for their degradation or elimination.

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Authors' contributions

GONÇALVES, E.V.: conceptualization; data curation; investigation; methodology; project administration; writing – original draft. ROQUEME, E.J.B.: formal analysis, methodology, validation, software, writing – original draft. MARTINS, M.D.: data curation; investigation; writing – original draft. CARAMORI, S.S.: conceptualization, supervision, validation, visualization, and writing – review and editing.

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