Wetlands in Brazil: classification, floristic composition and biological Nitrogen fixation

Áreas Úmidas no Brasil: classificação, composição florística e fixação biológica de Nitrogênio

Humedales en Brasil: clasificación, composición florística y fijación biológica de Nitrógeno

Abstract

Wetland ecosystems represent about 20% of South America, and are classified according to the flood regime, which also influences on vegetation. Despite the value of ecosystem services provided by this environment, those areas are close to eradication in several parts of Brazil. These environments are extremely fragile. Flooded areas are subject to nitrogen losses (N) by leaching, becoming dependent on the N increases from biological nitrogen fixation (BNF). However, little is known about this process on wetlands. Understanding the adaptive strategies of these microorganisms and plants is essential for the maintenance and preservation of these ecosystems. The objective of this work is to present a literature review discussing aspects of floristic composition, biological nitrogen fixation, and morphophysiological adaptations that occur in the rhizobium-leguminous system in wetlands. For the bibliographic survey, articles and other academic works relevant to the topic were selected, in order to enrich the proposed discussion.

Keywords: Adaptive strategies; Flooded areas; Legume; Nitrogen-fixing bacteria.
nitrógeno y adaptaciones morfofisiológicas que ocurren en el sistema rizobio-leguminosas en humedales. Para el levantamiento bibliográfico, se seleccionaron artículos y otros trabajos académicos relevantes al tema, con el fin de enriquecer la discusión propuesta.

**Palabras clave:** Áreas inundadas; Bacterias fijadoras de nitrógeno; Estrategias adaptativas; Legumbres.

1. Introduction

Wetland ecosystems involve all forms of flooded soils with vegetation, such as swamps, marshes, and mangroves (Mitsch & Gosselink, 2000). In South America, it is estimated that 20% of the total area is subject to flooding during periods of excessive rain. According to Costanza et al. (2014), the value of ecosystem services provided per unit area is 10-100 fold higher in wetlands when compared to dryland, making the understanding of the functioning of these areas vital (Neori & Agami, 2017).

The vegetation that occurs in these places can be classified, according to the flood regime, in alluvial and swamp forests (Silva et al., 2007), according to them, little is known about the species distribution patterns in flooded ecosystems, however, Marques et al. (2003), point out that, as they present a more selective, homogeneous and stable environment, swamp forests tend to have fewer species.

Despite their great importance as regulators of erosion processes, stabilization of margins, and promotion of nutrient cycling, the areas subject to flooding (permanent or temporary) are close to eradication in several parts of Brazil. These environments are extremely fragile and tend to have acidic soils, with slow drainage and high levels of organic matter, mainly due to the reduction in microbial activity caused by the absence or low concentration of oxygen.

Flooded areas are subject to nitrogen losses (N) by leaching, becoming dependent on the N increases from biological nitrogen fixation (BNF), where the rhizobium-leguminous association is the main contributor (Loureiro et al., 1998). However, legumes are, mainly, sensitive to flooding, producing additional mechanisms to increase the oxygen supply, making it possible to maintain the BNF process.

Little is known about BNF and nodular diazotrophic bacteria colonizing plant species from tropical forests (Cassetari, 2011). Krishnam et al. (2019), observed the formation of nodules in Sesbania herbacea (Mill.) under flooding conditions, the authors reported that flooding increases the number of nodules on Sesbania roots, and a BLAST analysis revealed a 100% sequence homology to 16S ribosomal RNA of Neorhizobium huautlense. Another studied by Brasil et al. (2016) about the influence of flood areas on the number of diazotrophic bacteria from pasture grasses showed that the presence of Azospirillum and Herbaspirillum presented high number in grasses Hymenachne amplexicaulis of permanent flood areas. Understanding the strategies used to increase the availability of N in flooded soils is essential for the maintenance and preservation of these ecosystems. The objective of this work is to present a literature review discussing aspects of floristic composition, biological nitrogen fixation, and morphophysiological adaptations that occur in the rhizobium-leguminous system in wetlands.

2. Methodology

In order to achieve the objectives, a systematic review was done. A systematic review is important because allows different points of view of the same subject. In addition, the use of this methodology identifies gaps in the literature, providing trustworthy data for future studies. The data collection followed four steps: planning, research, sorting, and analysis of the content (Figure 1). Three data bases were used in this study: Scielo, Scopus, and Google Scholar, using the index terms: wetlands in Brazil, rhizobium, sustainability, and floristic composition.
The inclusion criteria were based on the relevance of the paper for the research, as well as the language, selecting only paper in English or Portuguese. However, as exclusion criteria it was excluded the papers with a lot of similarity, being chosen those published most recently. Also, to select the papers, first it was reviewed the titles and abstracts, and articles that did not meet the objectives of the present study were excluded. Then, the full text of the articles previously selected was read, and if there was lack of relevant information, the paper was excluded.

Scanning and Skimming were the techniques used to read and select the texts. Skimming allows the reader to get a general overview of the material, and Scanning, on the other hand, permits to find specific information. Finally, once the papers were selected, the analysis of the content was done to summarize the main ideas, to fulfill the gaps, and answer the questions to achieve the literature review objectives.

3. Results and Discussion

Wetlands in Brazil

Wetlands cover approximately 20% of the Brazilian territory (Junk et al., 2015), and more than 90% are in the interior of the country, as a result of high precipitation and flat relief. In 1971, the first international wetland convention was held in Ramsar, Iran, which is characterized by being the first worldwide meeting related to the enhancement of these environments (Siman Gomes & Magalhaes Júnior, 2018)

In 1993, Brazil became a member of the Ramsar convention, becoming responsible for surveying, classifying, and promoting management and conservation studies in its wetlands. However, little progress was noted in the application of these criteria, making, according to the National Institute of Wetlands (INAU), the ecological and environmental functions of these Brazilian ecosystems little known and undervalued in legal and socio-political terms.

Due to its territorial extension, Brazil has a wide variety of wetlands, according to Junk (2011), the heterogeneity is related to variations in the rainfall regime, creating a mosaic of different types of wetlands. Junk et al. (2015) indicate that there are about 111 terminologies found in the Brazilian legal system. Table 1 shows those that stand out according to the author.
Table 1: Popular names for the different types of Brazilian wetlands and their characterization.

| Popular name                          | Region                        | Characterization                                                                                                                                 |
|---------------------------------------|-------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------|
| Baixada litorânea (Restinga)          | Coastal area                  | Bodies of shallow water and swamps between dunes on the coast, outcropping groundwater, with aquatic and palustres macrophytes, even forested. |
| Banhado                               | South of Brazil               | General designation of wetlands in Rio Grande do Sul.                                                                                           |
| Brinquihal/ Brejo                      | Paraná                        | Lowland forest. A little specific name for waterlogged areas.                                                                                   |
| Buritizal                             | Brazil                        | Soaked areas covered with buritis (*Mauritia flexuosa*).                                                                                        |
| Campina/ Campinarama                  | Central Amazon                | Sandy areas with periodically soaked soils, covered by hydromorphic savanna vegetation.                                                        |
| Carnaubal                             | Coastal area                  | Fresh water-soaked areas, dominated by the Carnaubá palm (*Copernicia prunifera*) and palm trees.                                              |
| Caxetal                               | Southeast/south               | Peat / muddy forest with the dominance of *Tabebuia cassinosides* Lam.                                                                       |
| Chavascal                             | Amazon                        | Permanently soaked area, covered with highly flood resistant forest.                                                                           |
| Estuário                              | Brazil                        | Coastal wetlands characterized as the final areas of rivers or lakes with strong influence of tides and saline water.                         |
| Igapó                                 | Central Amazon                | Floodable area along rivers of black and clear water, poor in nutrients.                                                                      |
| Lagoa                                 | Brazil                        | Permanent or temporary bodies of water throughout the national territory.                                                                      |
| Laguna Costeira                       | Coastal area                  | Bodies of water along the coast, usually of salinity and vegetation variable.                                                                   |
| Lavrados                              | Roraima                       | Savannah areas with lakes, swamps and footpaths dominated by *Mauritia flexuosa*.                                                              |
| Manguezaí                             | Coastal area                  | Coastal ecosystem, which occupies muddy, clayey or sandy sedimentary deposits up to the upper limit of the equinocial high tides.            |
| Mata Ciliar                           | Brazil                        | Wetland around bodies of water.                                                                                                               |
| Mata ripária/ Mata de galeria          | Brazil                        | Periodically flooded forest along rivers.                                                                                                       |
| Mata turfosa/ paludosa                | Southeast/ South              | It is characterized by a very particular floristic and structure, differentiating itself from other forest formations by its species capable of germinating and growing under conditions of water saturation. |
| Nascente/ Olho d’água                 | Brazil                        | Areas for discharging water from groundwater or subsurface water.                                                                             |
| Pântano                               | Brazil                        | A specific name for waterlogged areas.                                                                                                          |
| Restinga                              | Coastal area                  | Bodies of shallow water and swamps between dunes on the coast, outcropping groundwater.                                                      |
| Turfeira                              | South Brazil                  | Small humid areas located in high altitude areas or on the coastal plain with a high concentration of decomposing organic matter and low pH (acidic waters). |
| Vargem                               | Brazil                        | Any type of periodically flooded area.                                                                                                          |
| Varjão                                | Mato Grosso, Tocantins, Goiás | Very large floodplain in savanna areas.                                                                                                         |
| Várzea                                | Central Amazon                | Floodable area along white water rivers of Andean origin, rich in nutrients.                                                                  |
| Várzea                                | Other regions                 | Any type of periodically flooded area.                                                                                                          |
| Vereda                                | Brazilian cerrado             | Permanently humid area, covered by grassy herbaceous vegetation.                                                                               |

Source: adapted from Junk et al. (2015).

Concerning Brazilian legislation there are still gaps related to this ecosystem. Its definition in the Forest Code excludes permanent Wetlands and does not clarify whether wetlands are only those flooded by watercourses (Siman Gomes & Magalhães Junior, 2017), defining it as wetlands and land surfaces flooded natural and periodically, originally covered by forests or other forms of vegetation adapted to flooding (BRASIL, 2012). Wetlands can be classified according to their biological, ecological, physical, chemical, hydrological, or geomorphological attributes. Understanding the objectives and grouping the wetlands into similar units is essential to have a better description, assessment, and comparison of the wetlands.
for research and development of conservation programs and environmental impact assessment (Siman Gomes & Magalhães Junior, 2018).

**Wetland Classification Systems in Brazil**

There are several instruments used for the classification of wetlands internationally. When classified in systems there are two trends, horizontal and vertical (hierarchical). Horizontal divide habitats into classes or types, while hierarchical are separate into levels, from general to specific characteristics (Siman Gomes & Magalhães Junior, 2018). According to Junk et al. (2021), the parameters used to classify these habitats should address the specific, well-defined characteristics of each macrohabitat, hence, allowing the establishment of a databank with the necessary information.

In Brazil, institutions such as the National Institute of Wetlands (INAU) and the laboratory of ecology and conservation of aquatic ecosystems at the University of Valley of Rio dos Sinos (UNISINOS), carried out research to delimit and classify some of the large wetlands and their main habitats (Junk et al., 2013), however, these works are presented only from an ecological perspective, not incorporating geomorphological criteria. Because of the lack scientifically based classification system in Brazil, Maltchik et al. (2004), proposed the first hierarchical classification of wetland areas in the Rio Grande do Sul, presenting five levels: System, subsystem, type, class, and subclass. In 2012, Junk et al. (2012), through INAU, proposed a system for the entire Brazilian territory, to assist in the formation of a national wetland policy. It was based on three hierarchical levels: systems, units defined by hydrological factors (Subsystem, orders, and suborders) and units defined by higher plants (Class, subclass, and macro habitats).

Therefore, the Brazilian classification proposal does not incorporate geomorphological criteria or hydrogeomorphological classes (HGM), however, Junk et al. (2015), evaluate that these parameters are more useful from a scientific point of view, with no contribution to the political discussion in the management of Brazilian wetlands.

Brazil, although it has some classification systems, is weakened, since some regions remain absent, as is the case of Minas Gerais, where a classification system for Wetlands has not yet been conceived (Siman Gomes & Magalhães Junior, 2018).

**Floristic composition in wetlands**

The areas subject to flooding occur predominantly on the shores of rivers and lakes, or outcrops of the groundwater (Silva et al., 2007), and their floristic pattern is determined by the climate, edaphic factors, surrounding vegetation as a source of propagule (Rodrigues & Shepherd, 2000), anthropic actions and periodicity, duration and depth of flooding (Junk, 1993).

Pantanal is the largest tropical wetland in the world and the most important in Brazil. According to Pott et al. (2011), considering only the Pantanal plain there are approximately 2000 listed species of local flora, most belonging to the Fabaceae (240), Poaceae (212) and Malvaceae (98), with emphasis on the genera *Paspalum* (35), *Cyperus* (29) and *Ipomoea* (24).

Swampy forests are those that remain permanently flooded, being, according to Teixeira and Assis (2011), environments that have low species diversity and high local densities. Silva et al. (2007), characterize the floristic composition of these areas as homogeneous. The lower number of species in the swampy forests can be explained by the more selective, homogeneous, and stable environment, with constant flooding throughout the year (Marques et al., 2003). Table 2 shows the main families found in these environments, according to some surveys.
Table 2: Families found in swampy forests in Minas Gerais (MG) and coastal regions of Brazil.

| Study area                                      | Number of identified individuals | Main families | Author                  |
|------------------------------------------------|---------------------------------|---------------|-------------------------|
| Swampy forest in the south of MG                | 110 species 50 families         | Myrtaceae     | Loures et al., 2007     |
| Swampy habitat in MG                           | 99 species 35 families          | Fabaceae      | Rocha et al., 2005      |
| Swanpy restinga forest in the coastal plain of | 38 species 22 families          | Myrtaceae     | Santos-Junior et al., 2015 |
| southern brazil                                 |                                 | Meliaceae     |                         |

Source: Authors (2022).

Alluvial forests, on the other hand, are vegetations subject to temporary flooding (Silva et al., 2007), these environments are distributed over the most different areas of the country, presenting remarkable compositions of biodiversity (Ab'saber, 2001). Table 3 indicates the families that stand out in the floristic composition of alluvial areas in some studies.

Table 3: Families found in alluvial forest in Brazil.

| Study area                                      | Main families | Authors                          |
|------------------------------------------------|---------------|----------------------------------|
| Alluvial forests of southern and southeastern Brazil | Myrtaceae Fabaceae | Silva et al., 2007               |
| Paraíba do Sul, São Paulo, Brazil               | Myrtaceae Fabaceae | D’Oráazio and Catharino, 2013    |
| Survey of the state of São Paulo                | Fabaceae Myrtaceae Rubiaceae | Bertoni and Martins, 1987 Bertani et al., 2001 Teixeira and Assis, 2005 Aquino, 2006 Teixeira e Assis, 2009 |

Fonte: Autors (2021).

Riverside is another type of flooded environment, with a characteristic vegetal formation associated with water bodies (Oliveira-Filho, 1994), being considered areas that have an important role as corridors of plant and animal gene flow (Marinho Filho and Gastal, 2004). A floristic study carried out by Lacerda et al. (2010), in riverside areas of the caatinga, indicated a total of 91 species being Fabaceae, Euphorbiaceae, and Rubiaceae the families with the largest number of individuals and genera. The authors found that the greatest floristic identity is mainly related to the geographical distance and the characteristics of land use.

Considering the impacts of global climate change, the importance of wetlands tends to increase, making studies on the characterization of these ecosystems increasingly necessary.

Legumes in wetlands

Leguminosae (Fabaceae) is the third-largest family of Angiosperms, including about 760 genera and 19,500 species (Yahara et al., 2013), covering a great diversity of growth patterns (Doyle and Luckow, 2003). The height and duration of the periodic flooding induce the appearance of changes in the ecophysiological behavior of trees that colonize flooded areas, making it possible to adapt to conditions of oxygen scarcity for long periods (Wittmann et al., 2006).
These tree species survive in a dormant state and may also show vigorous growth in the flooded phase (Parolin et al., 2004). However, legumes are generally sensitive to flooding, making it a limiting factor for their growth (Loureiro et al., 1998). Table 4 shows the main legume species found in wetlands in the south and southeast regions of the country.

### Table 4: Legumes in wetlands in the south and southeast regions of Brazil.

| Species                                      | Authors                                      |
|----------------------------------------------|----------------------------------------------|
| *Acacia polyphylla* DC                       | Silva et al., 2007; Campos and Landgraf, 2001|
| *Apuleia leiocarpa* (Vogel) Macbr            | Silva et al., 2007                           |
| *Copaifera langsdorfii* Desf                 | Silva et al., 2007; Campos and Landgraf, 2001|
| *Dalbergia frutescens* (Vell.) Britton       | Silva et al., 2007                           |
| *Dalbergia miscolobium* Bent                  | Campos and Landgraf, 2001                     |
| *Erythrina falcata* Bent                     | Silva et al., 2007; Loures et al., 2007      |
| *Inga marginata* Wild                        | Silva et al., 2007                           |
| *Inga uruguaensis* Hooker at Arnott          | Campos and Landgraf, 2001                     |
| *Inga vulpina* Mart ex. O. Benth             | Loures et al., 2007                          |
| *Inga sp.* Hooker at Arnott                  | Campos and Landgraf, 2001                     |
| *Inga vera* Wild                             | Silva et al., 2007                           |
| *Lonchocarpu guillemineaus* (Tul.) Malme     | Loures et al., 2007                          |
| *Lonchocarpu mucilbergianus* Hassl           | Campos and Landgraf, 2001                     |
| *Machaerium hirtum* (Vell) Stellfeld         | Silva et al., 2007                           |
| *Machaerium minutiflorum*                    | Loures et al., 2007; Silva et al., 2007      |
| *Machaerium nyctitans* (Vell.) Benth         | Loures et al., 2007                          |
| *Machaerium paraguariense* Hassl             | Silva et al., 2007                           |
| *Parapiptadenia rigida* (Benth) Brenan       | Silva et al., 2007; Campos and Landgraf, 2001|
| *Pelogyne angustiflora* Ducke                | Campos and Landgraf, 2001                     |
| *Platycamus regnelli* Vog.                   | Campos and Landgraf, 2001                     |
| *Pterocarpu cidicens* Vog.                   | Campos and Landgraf, 2001                     |
| *Sclerolobium* sp.                           | Campos and Landgraf, 2001                     |
| *Sweetia fruticosa* Spreng                   | Campos and Landgraf, 2001                     |

Fonte: Autors (2021).

**Effects of flooding on soil and plants**

Flooding interferes with a series of physical-chemical and biological processes, deeply influencing soil quality. Among the changes, the following stand out: decreased gas exchange between the soil and the air; accumulation of gases such as N₂, CO₂ and H₂, the production of hydrocarbons, phenolic compounds, alcohols, and volatile fatty acids, due to anaerobiosis; increase in pH in acidic soils and its reduction in alkalis, and a significant decrease in redox potential (Ponnamperuma, 1984; Gambrell et al., 1991).

The excess of water in the soil, resulting from permanent or temporary flooding, influences the species composition (Jackson and Colmer, 2005), and the stress imposed by water saturation in the soil has a highly selective character (Medri et al., 2012). According to Joly (1991), in most species, success for survival is related to morphological, physiological, and anatomical adaptations.
Physiological adaptations in response to flooding consist of greater stomatal resistance, drop-in photosynthesis, and hydraulic conductance of the root, in addition to reduced translocation of photoassimilates (Striker, 2012 and Parent et al., 2008; Kolowski, 1997). Regarding morphological or structural adaptations, there is the formation of lenticels, aerenchyma, adventitious roots, pneumatophore, sapopemas, biological nitrogen fixation, and others (Parolin, 2012).

Also noteworthy for species underwater saturation conditions are phenological and reproductive adaptations, the former being related to leaf loss, fruit ripening, and seed release, and the latter associated with submersion tolerance, seed dormancy, and immediate germination (Parolin, 2012; Kolowski, 1997).

Pires et al. (2002), evaluating the effects of flooding on the morphophysiological characteristics of soybeans, observed that the main changes were in the roots, where was noted the death of the main root, the growth of lateral roots, and the appearance of adventitious roots, in addition to the decline of the levels of nutrients in the leaves.

**Biological Nitrogen Fixation in wetlands**

Nitrogen, even though it is one of the elements in greatest concentration in the atmosphere (78%), is found in a form not available (N₂) for most living beings, including plants. N₂-fixing bacteria, known as diazotrophic bacteria, can fix N₂ directly from the atmosphere through the biological nitrogen fixation process (BNF). (Cleveland et al., 1999; Boddey et al., 2000).

Plants in symbiosis with diazotrophic bacteria can occupy different ecosystems, adapting to the wide variety of environmental stress. Some species of actinorhizal plants are very well adapted to wetlands, arid regions, contaminated soils, extreme pH and high salinity, and, due to these properties, some of these plants are pioneers that colonize disturbed areas (Santi et al., 2013). However, soil flooding impairs the nodulation of legumes and inhibits N₂ fixation in previously formed nodules (Jackson, 1985).

The BNF process is closely related to edaphoclimatic factors, and wetlands are often subject to annual net nitrogen losses via leaching and are therefore largely dependent on the biological fixation process to ensure the entry of N into the system (Loureiro et al., 1998). Therefore, as highlighted by Hu et al. (2021), nitrogen is a common limiting nutrient for plant yield in wetlands. Consequently, most legumes that associate with diazotrophic bacteria in flooded regions have developed additional mechanisms that increase the supply of oxygen to their nodules, thus maintaining the capacity to fix nitrogen (Loureiro et al., 1998).

Legumes in flood conditions may have a limited supply of O₂ for root nodules. In these environments, according to Ladha et al. (1992), stem nodulation is an advantage. Loureiro (1994), states that the stem nodules receive oxygen via aerenchyma, which allows the diffusion of gases. James et al. (2001), studying flooding-tolerant legume symbioses from the Brazilian Pantanal, observed nodules on the stem of *Discolobium leptophyllum*.

Krishnam et al. (2019), observed that the nodules of *Sesbania herbacea* grown in flooded soils were larger and more numerous concerning the ones of plants grown in dry soils. The same effect was seen by Kanu and Dakora (2015), in a study with *Psoralea pinnata* (L.), which in flooded areas presented nodules with greater area and volume when compared to non-flooded regions. These authors also observed that the nodules had six components: lenticels, periderm, cortex (internal, middle, and external), and infection by bacteria in the central region of the spinal cord.

### 4. Final Considerations

Although wetlands represent about 20% of Brazilian territory, these ecosystems are unprotected by law, mainly due to differences in their concepts and classification criteria. Wetlands are present in all biomes of the country. However, some states
still lack a specific classification system that serves as a basis to ensure the preservation and maintenance of these environments.

Studies focused on these ecosystems are essential given the importance of environmental services, which have immeasurable value and ensure the sustainability of processes. The relationship between rhizobium and leguminous plants has been studied in the most diverse environments. Therefore, for wetlands, little is known about this process. The biological nitrogen fixation is considered the main mean of entry of nitrogen into these systems being so, knowing and understanding the adaptive strategies and identifying the species involved is necessary for the maintenance of biodiversity and preparation of management and conservation programs for wetlands.

In a nutshell, further studies are needed for Brazil to create more effective environmental laws, and to encourage research related to the subject and develop specific classification systems for the states to comply with the agreement signed by the Ramsar Convention. In addition, future works aiming to understand how plants and microorganisms adapt in those ecosystems are essential to create public awareness about the importance of conserving these areas.

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