











Periphytic algal flora of the lower Doce river basin after ore tailings flow, Espírito Santo State, Brazil: Sampling design and methods

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ABSTRACT - (Periphytic algal flora of the lower Doce river basin after ore tailings flow, Espírito Santo State, Brazil: Sampling design and methods). The present work aims to describe the sampling design and the methods used in the series “Periphytic algal flora of the Lower Doce River basin (Espírito Santo State, Brazil) after ore tailings flow”. This series will present the biodiversity of periphytic algae registered during the Aquatic Biodiversity Monitoring Program, Environmental Area I, carried out in the Lower Doce River basin three years after the passage of the ore tailings from Fundão dam (Mariana, Minas Gerais). Samples from the periphytic community were collected monthly, between October/2018 and February/2020, at five sampling stations in lotic environments and seven in lentic environments of the basin. These samples were fixed and the taxa will be identified and described, following specific methods. These taxa will be presented in sequential fascicles, split according to their taxonomic and morphological classification. More than 900 taxa have been recorded in this survey. This is the first taxonomic study about inland algae carried out in the Lower Doce River basin. **Keywords:** epilithon, epiphyton, freshwater diversity, lakes, rivers

RESUMO - (Flora de algas perifíticas da bacia do baixo rio Doce, após a passagem de rejeitos de minério, Estado do Espírito Santo, Brasil: Delineamento amostral e métodos). O presente trabalho tem como objetivo descrever o delineamento amostral e os métodos utilizados na série “Flora de algas perifíticas da bacia do baixo rio Doce, após a passagem de rejeitos de minério, Estado do Espírito Santo, Brasil”. Esta série apresentará a biodiversidade de algas perifíticas registradas durante o Programa de Monitoramento da Biodiversidade Aquática, Área Ambiental I, realizado na bacia do baixo rio Doce três anos após a passagem do rejeito de minério da barragem de Fundão, Mariana, Estado de Minas Gerais. Foram coletadas mensalmente mensalmente, entre outubro/2018 e fevereiro/2020, amostras da comunidade perifítica em cinco estações amostrais em ambientes lóticos e sete em ambientes lênticos da bacia. Essas amostras foram fixadas e os táxons serão identificados e descritos, seguindo métodos específicos. Esses táxons serão apresentados em fascículos sequenciais, divididos de acordo com sua classificação taxonômica e morfológica. Mais de 900 táxons foram registrados no levantamento. Este é o primeiro trabalho taxonômico sobre algas continentais realizado na bacia do baixo rio Doce.

Palavras-chave: epilítion, epifítion, diversidade de água doce, lagos, rios

Introduction

Phycofloristic studies are important for the local and regional biodiversity knowledge (Bicudo 2020), for species autecology, and also to correct identification of these organisms when they are used in biomonitoring programs (Hamada & Ferreira-Kepler 2012). Menezes et al. (2015) recorded 4,747 species of algae and cyanobacteria for Brazil. This number is increasing even more with several study efforts, such as the algae flora of streams in the Parnaíba River basin (Auricchio et al. 2019), checklist of the *Cosmarium* in Brazil (Biolo & Bicudo 2018), floristic survey of urban parks (D’Alessandro & Nogueira 2017), and descriptions of new species (Ramos et al. 2017, Lehmkuhl et al. 2019, Ramos et al. 2019, Zorzal-Almeida et al. 2020). One of the most relevant studies about Brazilian microalgae flora is the survey of the ‘Algal Flora of the Parque Estadual

das Fontes do Ipiranga (PEFI)’, which started in 1962 and registered 869 taxa in 42 fascicles (Bicudo 2020).

Algal flora of inland waters in Espírito Santo State, considering peer-reviewed works with illustration and description, is represented by Delazari-Barroso et al. (2007), recording phytoplankton community (except diatoms) from a dam, and the description of new cyanobacteria species (Senna et al. 1999). Thus, considering the high density of aquatic ecosystems in the State (Barroso 2007), the knowledge about the continental algae flora is practically null. This fact is aggravated by the threats to aquatic ecosystems that are increasing in frequency (Dudgeon et al. 2006, Reid et al. 2019), which lead to a decreasing in algal diversity (Wengrat et al. 2018) before they could be observed by researchers. For Espírito Santo State, it can be mentioned the case of Fundão dam rupture, in 2015, which released more than 43 million square meters of iron ore tailings in Rio Doce basin waters (Renova 2016). The

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tailings load drastically affected the Doce river, which has been historically contaminated by mining, causing a substantial increase in the loading of suspended sediments and transported various dissolved metals, such as iron and aluminium (Hatje et al. 2017). Besides the contamination of the main channel of Doce River, some adjacent lake ecosystems in the basin were also affected by the tailings. Although data about the effects in periphytic community integrity has not yet been published, it is important to highlight that surveys of the flora of microalgae (both periphyton and phytoplankton) were non-existent. For this reason, the real species pool in the region before the tailings pass is unknown, making it difficult to understand the real impact on the algae flora.

Works with periphytic algae community in Espírito Santo State are restricted to ecological approaches (e.g. Cavati & Fernandes 2007, Almeida & Fernandes 2013). This community is an important component of aquatic biodiversity (Lowe & Pan 1996) playing important roles in ecological processes, such as trophic chain (Vadeboncoeur & Steinman 2002) and biogeochemical cycles (Hagerthey et al. 2011), and it is considered an important bioindicator of the integrity of aquatic ecosystems (Lobo et al. 2014). Then, it is crucial to know which species make up the current regional pool of the Lower Doce River basin. This paper aims to describe the sample design and methods for the development of the series “Periphytic algal flora of the Lower Doce River basin (Espírito Santo State, Brazil) after ore tailings flow”. This series will be divided into fascicles according to taxonomic groups found in the periphytic community of the lentic and lotic aquatic ecosystems of the Lower Doce river studied in the Aquatic Biodiversity Monitoring Program, Environmental Area I, Periphyton

project, carried out by the research group “Rede Rio Doce-Mar”, starting to unravel their algae biodiversity.

Material and Methods

Study area - Lower Doce River basin is fully located in the State of Espírito Santo, southeast Brazil (41°30' to 39°30'W and 19°30' to 20°30'S) in the Tertiary plateaus formed by Barreiras Formation and the Quaternary Coastal Plain (Salinas et al. 2020). Its drainage area covers about 11,921 km² and contains about 90 smaller lakes, including the second largest Brazilian freshwater lake (Juparanã) (Barroso et al. 2012). According to the Köppen classification, region climate is Aw (humid tropical with dry winter) (Nóbrega et al. 2008) and seasonality can be divided into dry (April to September) and rainy (October to March) seasons (RRDM 2019), with 1,123 mm of average annual rainfall (Salinas et al. 2020). Lakes and lagoons in Lower Doce River, due to their formation, are often connected with Doce River in a bidirectional flow that varies according to the level of the river. Lower Doce River riparian area is highly degraded, with a large portion with more than 80% deforested (Pires et al. 2017), one of the causes of the intense silting up of rivers. Discharge of untreated domestic sewage in most municipalities in the basin has also caused the degradation of the water quality of the Doce River and its tributaries (ANA 2016).

Sampling design and material preparing - Samples were collected in four sampling sites in shallow lakes (Areão - E23, Areal - E24, and Monsarás - E25 and E25a), three in deep lakes (Limão - E18, Nova - E19, and Juparanã - E20), four in Doce River (E0, E21, E22, E26), and one in its affluent (Guandu river - E17) (figure 1).

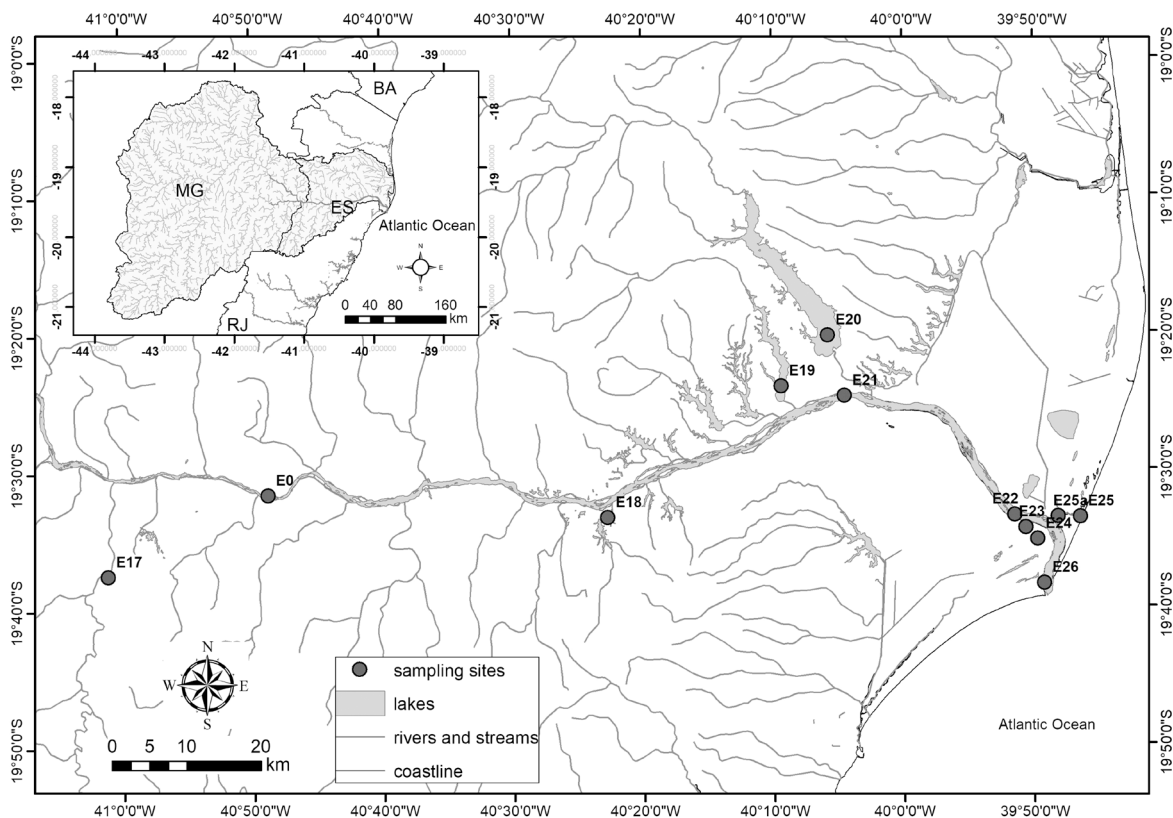


Figure 1. Sampling stations in the lower Doce river basin, Espírito Santo State, Brazil. Development by Gilberto F. Barroso.

Our choices of the sampling sites were based on environmental kind (river, lake, lagoon) and the possible impact of the ore tailing. Samplings were carried out monthly between October 2018 and February 2020 (except for October 2019 due to logistic problems). Periphytic material was collected from different substrates in littoral zone, such as pebbles and macrophytes. Whenever possible, the same kind of substrate was collected at each sampling station throughout the campaign. Substrates were transported to the laboratory in refrigerated recipients containing a small amount of distilled water to keep samples humid. At the laboratory, periphytic material was scrapped from the substrates with a smooth bristle brush and small jets of distilled water (following recommendations in Ferragut *et al.* 2013). All biological material was fixed and preserved with 3-5% formalin solution. For diatoms, organic materials were removed using hydrogen peroxide (H₂O₂ 35%) and hydrochloric acid (HCl 10%), following Battarbee *et al.* (2001), and permanent slides were mounted using Naphrax® (IR = 1.73) as the inclusion medium. Samples were deposited at the Herbário VIES at Federal University of Espírito Santo. We created for each herbarium sample a code that will be used to represent all information about the deposited sample (such as locality, sampling date, herbarium number, and substrate), as shown in table 2. For this purpose, we considered that all periphytic samples were collected in Espírito Santo State, Brazil.

Conductivity and pH were measured in the field (YSI Horiba U-53 or Exo2 multiparameter) and superficial water samples were taken to determine total iron (Inductively Coupled Plasma Atomic Spectrometry with detection by Spectrometry of Masses - ICP-MS), total phosphorous (Valderrama 1981), and chlorophyll-a (fluorimetric method; Barroso & Littlepage 1998). Trophic state index was calculated following Lamparelli (2004). For more details about abiotic variables, see RRDM (2019). These variables were selected since they represent ecosystem metabolism, eutrophication, and metal content in water.

Taxonomic and ecological analysis - For non-diatom species, a minimum of five non-permanent slides were prepared for each sample for the taxonomic survey and specimens were observed and photographed.

For diatoms species, the analysis were conducted in a full permanent slide per sampling site. Observed taxonomic features and the bibliography used in identification will be described in each fascicule, as it is specific to each algae group. Autecology will be based on the environmental variables range in which the species were found. Yet, taxa distribution was assess using the frequency of occurrence and will be calculated to classify them according to the categories in Matteucci & Colma (1982): Very Frequent: VF (> 70%), Frequent: F (≤ 70% and > 40%), Little Frequent: LF (≤ 40% and > 10%), and Rare: R (≤ 10%).

Results and Discussion

During the survey, 185 samples were collected along the Lower Doce River basin. Some samples could not be collected due to difficulty in accessing the sample station, or loss of the sample during transport. Sampling stations coordinates and environmental characterization (based on the variables used for the autecology of the species) are shown in table 1. The environments were characterized by a wide range of conductivity, total iron, and trophic states (from ultraoligotrophic to supereutrophic). Only the sampling stations of the lagoons could be classified as slight acid in some campaigns, while pH in other sampling stations never were less than 7. For more details on the ecosystem characteristics, see RRDM (2019). Table 2 shows the herbarium numbers and other related features and substrates from which periphytic material was scraped in each sampling site.

More than 900 taxa (~ 17 Classes) have already been registered and, “even though not all taxa identification have been finished”, we found that the most representative Classes were Bacillariophyceae (~300 taxa), Zygnematophyceae (~185 taxa), Cyanophyceae (~145 taxa), and Chlorophyceae (~125 taxa). After identification and description, these taxa will be grouped according to taxonomic groups (e.g., genus *Cosmarium*) and/or their morphology (e.g., coccoid Cyanophyceae), according to the number of species. Each group will compose a fascicule of the series about periphytic flora of the Lower Doce River basin, which will

Table 1: Sampling sites location and range of environmental conditions, Espírito Santo State, Brazil. Cond: conductivity. Fe: total iron. Trophic state - Ult: Ultraoligotrophic. Oli: oligotrophic. Mes: Mesotrophic. Eut: Eutrophic. Sup: Supereutrophic.

		Longitude	Latitude	μS cm ⁻¹		mg L ⁻¹	
E17	Guandu River	-41.018933	-19.625037	68-89	7.3-8.8	1.6-4.9	Ult-Mes
E0	Doce River, Itapina	-40.813257	-19.527360	56-121	7.1-8.5	0.8-3.0	Oli-Mes
E21	Doce River, Linhares	-40.070736	-19.406685	63-91	7.3-8.3	1.2-5.8	Oli-Eut
E22	Doce River, Povoação	-39.851833	-19.552639	53-105	7.3-8.5	1.5-7.1	Oli-Eut
E26	Doce River Mouth, Regência	-39.809056	-19.630433	52-1283	7.3-8.8	0.8-4.3	Oli-Eut
E18	Limão Lake	-40.378119	-19.557439	106-129	7.3-8.4	0.2-2.3	Oli-Sup
E19	Nova Lake	-40.155113	-19.399453	95-99	7.4-8.4	0.1-1.0	Oli-Eut
E20	Juparanã Lake	-40.095428	-19.337758	88-93	7.2-8.6	0.5-1.6	Oli-Sup
E23	Areão Lake	-39.843178	-19.571617	90-114	6.7-7.9	1.3-2.6	Oli-Mes
E24	Areal Lake	-39.828164	-19.585964	122-160	6.8-8.2	1.6-5.7	Oli-Sup
E25a	Monsarás Lake (near the bridge)	-39.801728	-19.558696	154-7067	6.4-7.8	1.7-4.3	Oli-Sup
E25	Monsarás Lake (near the beach)	-39.773448	-19.558964	483-4631	6.8-8.5	0.9-3.4	Oli-Sup

Table 2: Sample codes, herbarium numbers and attached information about locality, date, substrate and collector, lower Doce river basin after ore tailings flow, Espírito Santo State, Brazil.

Sample Code	Herbarium number	Sample	Locality	Date	Substrates	Collector
BRDE17A01	VIES Microalga 9800001	E17 out/18	Baixo Guandu	2018-X-23	Pebble	Zorzal-Almeida, S.
BRDE0A01	VIES Microalga 9800002	E0 out/18	Colatina	2018-X-24	<i>Ipomoea</i> sp.; Pebble	Zorzal-Almeida, S.
BRDE21A01	VIES Microalga 9800003	E21 out/18	Linhares	2018-X-24	<i>Eichhornia</i> sp.	Zorzal-Almeida, S.
BRDE26A01	VIES Microalga 9800004	E26 out/18	Linhares	2018-X-22	Poaceae; Dead branch	Zorzal-Almeida, S.
BRDE18A01	VIES Microalga 9800005	E18 out/18	Colatina	2018-X-23	<i>Eleocharis</i> sp.	Zorzal-Almeida, S.
BRDE19A01	VIES Microalga 9800006	E19 out/18	Linhares	2018-X-25	<i>Eichhornia</i> sp.	Zorzal-Almeida, S.
BRDE20A01	VIES Microalga 9800007	E20 out/18	Linhares	2018-X-25	Poaceae	Zorzal-Almeida, S.
BRDE23A01	VIES Microalga 9800008	E23 out/18	Linhares	2018-X-26	<i>Nymphaea</i> sp.	Zorzal-Almeida, S.
BRDE24A01	VIES Microalga 9800009	E24 out/18	Linhares	2018-X-26	<i>Nymphaea</i> sp.	Zorzal-Almeida, S.
BRDE25A01	VIES Microalga 9800010	E25 out/18	Linhares	2018-X-25	<i>Typha</i> sp.	Zorzal-Almeida, S.
BRDE25aA01	VIES Microalga 9800011	E25A out/18	Linhares	2018-X-25	<i>Nymphaea</i> sp.	Zorzal-Almeida, S.
BRDE17A02	VIES Microalga 9800012	E17 nov/18	Baixo Guandu	2018-XI-21	Pebble	Zorzal-Almeida, S.
BRDE00A02	VIES Microalga 9800013	E0 nov/18	Colatina	2018-XI-21	<i>Ludwigia</i> sp.; Pebble	Zorzal-Almeida, S.
BRDE21A02	VIES Microalga 9800014	E21 nov/18	Linhares	2018-XI-22	Apocynaceae; Poaceae	Zorzal-Almeida, S.
BRDE22A02	VIES Microalga 9800015	E22 nov/18	Linhares	2018-XI-19	Acanthaceae; <i>Mimosa</i> sp.	Zorzal-Almeida, S.
BRDE26A02	VIES Microalga 9800016	E26 nov/18	Linhares	2018-XI-20	Poaceae; <i>Talipariti pernambucensis</i>	Zorzal-Almeida, S.
BRDE18A02	VIES Microalga 9800017	E18 nov/18	Colatina	2018-XI-20	<i>Eleocharis</i> sp.	Zorzal-Almeida, S.
BRDE19A02	VIES Microalga 9800018	E19 nov/18	Linhares	2018-XI-22	<i>Eichhornia</i> sp.	Zorzal-Almeida, S.
BRDE20A02	VIES Microalga 9800019	E20 nov/18	Linhares	2018-XI-21	Poaceae	Zorzal-Almeida, S.
BRDE25A02	VIES Microalga 9800020	E25 nov/18	Linhares	2018-XI-19	<i>Typha</i> sp.	Zorzal-Almeida, S.
BRDE25aA02	VIES Microalga 9800021	E25A nov/18	Linhares	2018-XI-19	<i>Nymphaea</i> sp.	Zorzal-Almeida, S.
BRDE17A03	VIES Microalga 9800022	E17 dez/18	Baixo Guandu	2018-XII-12	Pebble	Zorzal-Almeida, S.
BRDE00A03	VIES Microalga 9800023	E0 dez/18	Colatina	2018-XII-12	<i>Ipomoea</i> sp.; Euphorbiaceae	Zorzal-Almeida, S.
BRDE21A03	VIES Microalga 9800024	E21 dez/18	Linhares	2018-XII-13	Acanthaceae; Poaceae	Zorzal-Almeida, S.
BRDE22A03	VIES Microalga 9800025	E22 dez/18	Linhares	2018-XII-12	Acanthaceae	Zorzal-Almeida, S.
BRDE26A03	VIES Microalga 9800026	E26 dez/18	Linhares	2018-XII-11	Amaranthaceae; <i>Talipariti pernambucensis</i>	Zorzal-Almeida, S.
BRDE18A03	VIES Microalga 9800027	E18 dez/18	Colatina	2018-XII-12	<i>Eleocharis</i> sp.	Zorzal-Almeida, S.
BRDE19A03	VIES Microalga 9800028	E19 dez/18	Linhares	2018-XII-13	<i>Eichhornia</i> sp.	Zorzal-Almeida, S.
BRDE20A03	VIES Microalga 9800029	E20 dez/18	Linhares	2018-XII-13	Poaceae	Zorzal-Almeida, S.
BRDE23A03	VIES Microalga 9800030	E23 dez/18	Linhares	2018-XII-11	Pontederiaceae	Zorzal-Almeida, S.
BRDE24A03	VIES Microalga 9800031	E24 dez/18	Linhares	2018-XII-11	<i>Eleocharis</i> sp.	Zorzal-Almeida, S.
BRDE25A03	VIES Microalga 9800032	E25 dez/18	Linhares	2018-XII-11	<i>Typha</i> sp.	Zorzal-Almeida, S.
BRDE25aA03	VIES Microalga 9800033	E25A dez/18	Linhares	2018-XII-11	<i>Nymphaea</i> sp.	Zorzal-Almeida, S.
BRDE17A04	VIES Microalga 9800034	E17 jan/19	Baixo Guandu	2019-I-15	Pebble	Zorzal-Almeida, S.
BRDE00A04	VIES Microalga 9800035	E0 jan/19	Colatina	2019-I-15	Poaceae; Pebble	Zorzal-Almeida, S.
BRDE21A04	VIES Microalga 9800036	E21 jan/19	Linhares	2019-I-15	Poaceae	Zorzal-Almeida, S.
BRDE22A04	VIES Microalga 9800037	E22 jan/19	Linhares	2019-I-14	Dead branch	Zorzal-Almeida, S.
BRDE26A04	VIES Microalga 9800038	E26 jan/19	Linhares	2019-I-16	Amaranthaceae; <i>Talipariti pernambucense</i>	Zorzal-Almeida, S.
BRDE18A04	VIES Microalga 9800039	E18 jan/19	Colatina	2019-I-17	<i>Eleocharis</i> sp.	Zorzal-Almeida, S.
BRDE19A04	VIES Microalga 9800040	E19 jan/19	Linhares	2019-I-17	<i>Eichhornia</i> sp.	Zorzal-Almeida, S.
BRDE20A04	VIES Microalga 9800041	E20 jan/19	Linhares	2019-I-16	<i>Eichhornia</i> sp.	Zorzal-Almeida, S.
BRDE23A04	VIES Microalga 9800042	E23 jan/19	Linhares	2019-I-17	<i>Eleocharis</i> sp.	Zorzal-Almeida, S.
BRDE24A04	VIES Microalga 9800043	E24 jan/19	Linhares	2019-I-16	<i>Limnocharis</i> sp.	Zorzal-Almeida, S.
BRDE25A04	VIES Microalga 9800044	E25 jan/19	Linhares	2019-I-14	<i>Typha domingensis</i>	Zorzal-Almeida, S.
BRDE25aA04	VIES Microalga 9800045	E25A jan/19	Linhares	2019-I-14	<i>Nymphaea</i> sp.	Zorzal-Almeida, S.
BRDE17A05	VIES Microalga 9800046	E17 fev/19	Baixo Guandu	2019-II-13	Pebble	Zorzal-Almeida, S.

continue

Table 2 (continuation)

Sample Code	Herbarium number	Sample	Locality	Date	Substrates	Collector
BRDE00A05	VIES Microalga 9800047	E0 fev/19	Colatina	2019-II-13	Pebble; Dead Branch	Zorzal-Almeida, S.
BRDE21A05	VIES Microalga 9800048	E21 fev/19	Linhares	2019-II-14	Acanthaceae; Amaranthaceae	Zorzal-Almeida, S.
BRDE22A05	VIES Microalga 9800049	E22 fev/19	Linhares	2019-II-11	Acanthaceae	Zorzal-Almeida, S.
BRDE26A05	VIES Microalga 9800050	E26 fev/19	Linhares	2019-II-12	Amaranthaceae; <i>Talipariti pernambucense</i>	Zorzal-Almeida, S.
BRDE18A05	VIES Microalga 9800051	E18 fev/19	Colatina	2019-II-13	<i>Nymphaea</i> sp.	Zorzal-Almeida, S.
BRDE19A05	VIES Microalga 9800052	E19 fev/19	Linhares	2019-II-14	<i>Eichhornia</i> sp.	Zorzal-Almeida, S.
BRDE20A05	VIES Microalga 9800053	E20 fev/19	Linhares	2019-II-14	<i>Eichhornia</i> sp.	Zorzal-Almeida, S.
BRDE23A05	VIES Microalga 9800054	E23 fev/19	Linhares	2019-II-12	<i>Limncharis</i> sp.	Zorzal-Almeida, S.
BRDE24A05	VIES Microalga 9800055	E24 fev/19	Linhares	2019-II-12	<i>Nymphaea</i> sp.	Zorzal-Almeida, S.
BRDE25A05	VIES Microalga 9800056	E25 fev/19	Linhares	2019-II-11	<i>Typha domingensis</i>	Zorzal-Almeida, S.
BRDE25aA05	VIES Microalga 9800057	E25A fev/19	Linhares	2019-II-11	<i>Nymphaea</i> sp.	Zorzal-Almeida, S.
BRDE17A06	VIES Microalga 9800058	E17 mar/19	Baixo Guandu	2019-III-20	Pebble	Trancoso, M.S.
BRDE00A06	VIES Microalga 9800059	E0 mar/19	Colatina	2019-III-20	Poaceae	Trancoso, M.S.
BRDE21A06	VIES Microalga 9800060	E21 mar/19	Linhares	2019-III-21	<i>Culpea melvilla</i> ; <i>Polygonum</i> sp.	Trancoso, M.S.
BRDE22A06	VIES Microalga 9800061	E22 mar/19	Linhares	2019-III-18	<i>Culpea melvilla</i> ; Poaceae	Trancoso, M.S.
BRDE26A06	VIES Microalga 9800062	E26 mar/19	Linhares	2019-III-19	<i>Talipariti pernambucense</i> ; <i>Eichhornia</i> sp.	Trancoso, M.S.
BRDE18A06	VIES Microalga 9800063	E18 mar/19	Colatina	2019-III-20	<i>Nymphaea rudgeana</i>	Trancoso, M.S.
BRDE19A06	VIES Microalga 9800064	E19 mar/19	Linhares	2019-III-21	<i>Eichhornia</i> sp.	Trancoso, M.S.
BRDE20A06	VIES Microalga 9800065	E20 mar/19	Linhares	2019-III-21	Poaceae	Trancoso, M.S.
BRDE23A06	VIES Microalga 9800066	E23 mar/19	Linhares	2019-III-19	<i>Sagittaria</i> sp.	Trancoso, M.S.
BRDE24A06	VIES Microalga 9800067	E24 mar/19	Linhares	2019-III-19	<i>Nymphaea caerulea</i>	Trancoso, M.S.
BRDE25A06	VIES Microalga 9800068	E25 mar/19	Linhares	2019-III-18	<i>Typha domingensis</i>	Trancoso, M.S.
BRDE25aA06	VIES Microalga 9800069	E25A mar/19	Linhares	2019-III-18	<i>Nymphaea caerulea</i>	Trancoso, M.S.
BRDE17A07	VIES Microalga 9800070	E17 abr/19	Baixo Guandu	2019-IV-10	Pebble	Trancoso, M.S.
BRDE00A07	VIES Microalga 9800071	E0 abr/19	Colatina	2019-IV-10	Poaceae	Trancoso, M.S.
BRDE21A07	VIES Microalga 9800072	E21 abr/19	Linhares	2019-IV-11	Acanthaceae; Amaranthaceae	Trancoso, M.S.
BRDE22A07	VIES Microalga 9800073	E22 abr/19	Linhares	2019-IV-08	Acanthaceae	Trancoso, M.S.
BRDE26A07	VIES Microalga 9800074	E26 abr/19	Linhares	2019-IV-09	<i>Talipariti pernambucense</i> ; Poaceae	Trancoso, M.S.
BRDE18A07	VIES Microalga 9800075	E18 abr/19	Colatina	2019-IV-10	<i>Eleocharis</i> sp.	Trancoso, M.S.
BRDE19A07	VIES Microalga 9800076	E19 abr/19	Linhares	2019-IV-11	<i>Eichhornia</i> sp.	Trancoso, M.S.
BRDE20A07	VIES Microalga 9800077	E20 abr/19	Linhares	2019-IV-11	<i>Eichhornia</i> sp.	Trancoso, M.S.
BRDE23A07	VIES Microalga 9800078	E23 abr/19	Linhares	2019-IV-09	<i>Sagittaria</i> sp.	Trancoso, M.S.
BRDE24A07	VIES Microalga 9800079	E24 abr/19	Linhares	2019-IV-09	<i>Nymphaea</i> sp.	Trancoso, M.S.
BRDE25A07	VIES Microalga 9800080	E25 abr/19	Linhares	2019-IV-08	<i>Typha domingensis</i>	Trancoso, M.S.
BRDE25aA07	VIES Microalga 9800081	E25A abr/19	Linhares	2019-IV-08	<i>Nymphaea caerulea</i>	Trancoso, M.S.
BRDE17A08	VIES Microalga 9800082	E17 mai/19	Baixo Guandu	2019-V-15	Pebble	Souza, K.B.
BRDE00A08	VIES Microalga 9800083	E0 mai/19	Colatina	2019-V-15	Poaceae	Souza, K.B.
BRDE21A08	VIES Microalga 9800084	E21 mai/19	Linhares	2019-V-17	<i>Cuphea melvilla</i> ; Poaceae	Souza, K.B.
BRDE22A08	VIES Microalga 9800085	E22 mai/19	Linhares	2019-V-13	<i>Cuphea melvilla</i>	Souza, K.B.
BRDE26A08	VIES Microalga 9800086	E26 mai/19	Linhares	2019-V-13	<i>Talipariti pernambucense</i> ; <i>Eichhornia</i> sp.	Souza, K.B.
BRDE18A08	VIES Microalga 9800087	E18 mai/19	Colatina	2019-V-16	<i>Eleocharis</i> sp.	Souza, K.B.
BRDE19A08	VIES Microalga 9800088	E19 mai/19	Linhares	2019-V-16	<i>Eichhornia</i> sp.	Souza, K.B.
BRDE20A08	VIES Microalga 9800089	E20 mai/19	Linhares	2019-V-16	<i>Eichhornia</i> sp.	Souza, K.B.
BRDE23A08	VIES Microalga 9800090	E23 mai/19	Linhares	2019-V-14	<i>Sagittaria</i> sp.	Souza, K.B.
BRDE24A08	VIES Microalga 9800091	E24 mai/19	Linhares	2019-V-14	<i>Nymphaea caerulea</i>	Souza, K.B.
BRDE25A08	VIES Microalga 9800092	E25 mai/19	Linhares	2019-V-14	<i>Typha</i> sp.	Souza, K.B.
BRDE25aA08	VIES Microalga 9800093	E25A mai/19	Linhares	2019-V-14	<i>Nymphaea caerulea</i>	Souza, K.B.

continue

Table 2 (continuation)

Sample Code	Herbarium number	Sample	Locality	Date	Substrates	Collector
BRDE17A09	VIES Microalga 9800094	E17 jun/19	Baixo Guandu	2019-VI-12	Pebble	Souza, K.B.
BRDE00A09	VIES Microalga 9800095	E0 jun/19	Colatina	2019-VI-12	Poaceae	Souza, K.B.
BRDE21A09	VIES Microalga 9800096	E21 jun/19	Linhares	2019-VI-11	Poaceae	Souza, K.B.
BRDE22A09	VIES Microalga 9800097	E22 jun/19	Linhares	2019-VI-11	<i>Cuphea melvilla</i>	Souza, K.B.
BRDE26A09	VIES Microalga 9800098	E26 jun/19	Linhares	2019-VI-10	Poaceae	Souza, K.B.
BRDE18A09	VIES Microalga 9800099	E18 jun/19	Colatina	2019-VI-12	<i>Eleocharis</i> sp.	Souza, K.B.
BRDE19A09	VIES Microalga 9800100	E19 jun/19	Linhares	2019-VI-13	<i>Eichhornia</i> sp.	Souza, K.B.
BRDE20A09	VIES Microalga 9800101	E20 jun/19	Linhares	2019-VI-13	Poaceae	Souza, K.B.
BRDE23A09	VIES Microalga 9800102	E23 jun/19	Linhares	2019-VI-10	<i>Sagittaria</i> sp.	Souza, K.B.
BRDE24A09	VIES Microalga 9800103	E24 jun/19	Linhares	2019-VI-10	<i>Nymphaea</i> sp.	Souza, K.B.
BRDE25A09	VIES Microalga 9800104	E25 jun/19	Linhares	2019-VI-11	<i>Typha</i> sp.	Souza, K.B.
BRDE25aA09	VIES Microalga 9800105	E25A jun/19	Linhares	2019-VI-11	<i>Nymphaea</i> sp.	Souza, K.B.
BRDE17A10	VIES Microalga 9800106	E17 jul/19	Baixo Guandu	2019-VII-17	Pebble	Trancoso, M.S.
BRDE00A10	VIES Microalga 9800107	E0 jul/19	Colatina	2019-VII-17	Poaceae	Trancoso, M.S.
BRDE21A10	VIES Microalga 9800108	E21 jul/19	Linhares	2019-VII-16	Poaceae	Trancoso, M.S.
BRDE22A10	VIES Microalga 9800109	E22 jul/19	Linhares	2019-VII-16	<i>Cuphea melvilla</i>	Trancoso, M.S.
BRDE26A10	VIES Microalga 9800110	E26 jul/19	Linhares	2019-VII-15	<i>Talipariti pernambucense</i> ; Poaceae	Trancoso, M.S.
BRDE18A10	VIES Microalga 9800111	E18 jul/19	Colatina	2019-VII-17	<i>Eleocharis</i> sp.	Trancoso, M.S.
BRDE19A10	VIES Microalga 9800112	E19 jul/19	Linhares	2019-VII-18	<i>Eichhornia azurea</i>	Trancoso, M.S.
BRDE20A10	VIES Microalga 9800113	E20 jul/19	Linhares	2019-VII-18	Poaceae	Trancoso, M.S.
BRDE23A10	VIES Microalga 9800114	E23 jul/19	Linhares	2019-VII-15	<i>Sagittaria lancifolia</i>	Trancoso, M.S.
BRDE24A10	VIES Microalga 9800115	E24 jul/19	Linhares	2019-VII-15	<i>Eleocharis</i> sp.	Trancoso, M.S.
BRDE25A10	VIES Microalga 9800116	E25 jul/19	Linhares	2019-VII-16	<i>Typha domingensis</i>	Trancoso, M.S.
BRDE25aA10	VIES Microalga 9800117	E25A jul/19	Linhares	2019-VII-16	<i>Nymphaea caerulea</i>	Trancoso, M.S.
BRDE17A11	VIES Microalga 9800118	E17 ago/19	Baixo Guandu	2019-VIII-15	Pebble	Trancoso, M.S.
BRDE00A11	VIES Microalga 9800119	E0 ago/19	Colatina	2019-VIII-15	<i>Paspalum pilosum</i> ; Poaceae	Trancoso, M.S.
BRDE21A11	VIES Microalga 9800120	E21 ago/19	Linhares	2019-VIII-16	<i>Panicum aquaticum</i> ; Bambusoideae	Trancoso, M.S.
BRDE22A11	VIES Microalga 9800121	E22 ago/19	Linhares	2019-VIII-15	<i>Cuphea melvilla</i>	Trancoso, M.S.
BRDE26A11	VIES Microalga 9800122	E26 ago/19	Linhares	2019-VIII-12	<i>Talipariti pernambucense</i> ; Poaceae	Trancoso, M.S.
BRDE18A11	VIES Microalga 9800123	E18 ago/19	Colatina	2019-VIII-13	<i>Eleocharis interstincta</i>	Trancoso, M.S.
BRDE19A11	VIES Microalga 9800124	E19 ago/19	Linhares	2019-VIII-16	<i>Eichhornia azurea</i>	Trancoso, M.S.
BRDE20A11	VIES Microalga 9800125	E20 ago/19	Linhares	2019-VIII-14	Poaceae	Trancoso, M.S.
BRDE23A11	VIES Microalga 9800126	E23 ago/19	Linhares	2019-VIII-12	<i>Sagittaria lancifolia</i>	Trancoso, M.S.
BRDE24A11	VIES Microalga 9800127	E24 ago/19	Linhares	2019-VIII-12	<i>Eleocharis interstincta</i>	Trancoso, M.S.
BRDE25A11	VIES Microalga 9800128	E25 ago/19	Linhares	2019-VIII-13	<i>Typha domingensis</i>	Trancoso, M.S.
BRDE25aA11	VIES Microalga 9800129	E25A ago/19	Linhares	2019-VIII-13	<i>Nymphaea caerulea</i>	Trancoso, M.S.
BRDE17A12	VIES Microalga 9800130	E17 set/19	Baixo Guandu	2019-IX-18	Pebble	Trancoso, M.S.
BRDE00A12	VIES Microalga 9800131	E0 set/19	Colatina	2019-IX-18	<i>Paspalum pilosum</i> ; Poaceae	Trancoso, M.S.
BRDE21A12	VIES Microalga 9800132	E21 set/19	Linhares	2019-IX-17	<i>Panicum aquaticum</i> ; Bambusoideae	Trancoso, M.S.
BRDE22A12	VIES Microalga 9800133	E22 set/19	Linhares	2019-IX-17	<i>Cuphea melvilla</i>	Trancoso, M.S.
BRDE26A12	VIES Microalga 9800134	E26 set/19	Linhares	2019-IX-16	<i>Talipariti pernambucense</i> ; Poaceae	Trancoso, M.S.
BRDE18A12	VIES Microalga 9800135	E18 set/19	Colatina	2019-IX-18	<i>Eleocharis interstincta</i>	Trancoso, M.S.
BRDE19A12	VIES Microalga 9800136	E19 set/19	Linhares	2019-IX-19	<i>Eichhornia azurea</i>	Trancoso, M.S.
BRDE20A12	VIES Microalga 9800137	E20 set/19	Linhares	2019-IX-19	Poaceae	Trancoso, M.S.
BRDE23A12	VIES Microalga 9800138	E23 set/19	Linhares	2019-IX-16	<i>Sagittaria lancifolia</i>	Trancoso, M.S.
BRDE24A12	VIES Microalga 9800139	E24 set/19	Linhares	2019-IX-16	<i>Eleocharis interstincta</i>	Trancoso, M.S.
BRDE25A12	VIES Microalga 9800140	E25 set/19	Linhares	2019-IX-17	<i>Typha domingensis</i>	Trancoso, M.S.

continue

Table 2 (continuation)

Sample Code	Herbarium number	Sample	Locality	Date	Substrates	Collector
BRDE25aA12	VIÉS Microalga 9800141	E25A set/19	Linhares	2019-IX-17	<i>Nymphaea caerulea</i>	Trancoso, M.S.
BRDE17A13	VIÉS Microalga 9800142	E17 nov/19	Baixo Guandu	2019-XI-27	Pebble	Trancoso, M.S.
BRDE00A13	VIÉS Microalga 9800143	E0 nov/19	Colatina	2019-XI-27	<i>Paspalum pilosum</i>	Trancoso, M.S.
BRDE21A13	VIÉS Microalga 9800144	E21 nov/19	Linhares	2019-XI-28	<i>Panicum aquaticum</i> ; Bambusoideae	Trancoso, M.S.
BRDE22A13	VIÉS Microalga 9800145	E22 nov/19	Linhares	2019-XI-26	<i>Cuphea melvilla</i>	Trancoso, M.S.
BRDE26A13	VIÉS Microalga 9800146	E26 nov/19	Linhares	2019-XI-25	<i>Talipariti pernambucense</i> ; <i>Panicum aquaticum</i>	Trancoso, M.S.
BRDE18A13	VIÉS Microalga 9800147	E18 nov/19	Colatina	2019-XI-27	<i>Eleocharis interstincta</i>	Trancoso, M.S.
BRDE19A13	VIÉS Microalga 9800148	E19 nov/19	Linhares	2019-XI-28	<i>Eichhornia azurea</i>	Trancoso, M.S.
BRDE20A13	VIÉS Microalga 9800149	E20 nov/19	Linhares	2019-XI-28	<i>Panicum aquaticum</i>	Trancoso, M.S.
BRDE24A13	VIÉS Microalga 9800150	E24 nov/19	Linhares	2019-XI-25	<i>Eleocharis interstincta</i>	Trancoso, M.S.
BRDE25A13	VIÉS Microalga 9800151	E25 nov/19	Linhares	2019-XI-26	<i>Typha domingensis</i>	Trancoso, M.S.
BRDE25aA13	VIÉS Microalga 9800152	E25A nov/19	Linhares	2019-XI-26	<i>Nymphaea caerulea</i>	Trancoso, M.S.
BRDE17A14	VIÉS Microalga 9800153	E17 dez/19	Baixo Guandu	2019-XII-11	Pebble	Trancoso, M.S.
BRDE00A14	VIÉS Microalga 9800154	E0 dez/19	Colatina	2019-XII-11	<i>Paspalum pilosum</i>	Trancoso, M.S.
BRDE21A14	VIÉS Microalga 9800155	E21 dez/19	Linhares	2019-XII-10	<i>Panicum aquaticum</i> ; Bambusoideae	Trancoso, M.S.
BRDE22A14	VIÉS Microalga 9800156	E22 dez/19	Linhares	2019-XII-10	<i>Cuphea melvilla</i>	Trancoso, M.S.
BRDE26A14	VIÉS Microalga 9800157	E26 dez/19	Linhares	2019-XII-09	<i>Talipariti pernambucensis</i> ; <i>Panicum aquaticum</i>	Trancoso, M.S.
BRDE18A14	VIÉS Microalga 9800158	E18 dez/19	Colatina	2019-XII-11	<i>Eleocharis interstincta</i>	Trancoso, M.S.
BRDE19A14	VIÉS Microalga 9800159	E19 dez/19	Linhares	2019-XII-12	<i>Eichhornia azurea</i>	Trancoso, M.S.
BRDE20A14	VIÉS Microalga 9800160	E20 dez/19	Linhares	2019-XII-12	<i>Panicum aquaticum</i>	Trancoso, M.S.
BRDE24A14	VIÉS Microalga 9800161	E24 dez/19	Linhares	2019-XII-09	<i>Eleocharis interstincta</i>	Trancoso, M.S.
BRDE25A14	VIÉS Microalga 9800162	E25 dez/19	Linhares	2019-XII-10	<i>Typha domingensis</i>	Trancoso, M.S.
BRDE25aA14	VIÉS Microalga 9800163	E25A dez/19	Linhares	2019-XII-10	<i>Nymphaea rudgeana</i>	Trancoso, M.S.
BRDE17A15	VIÉS Microalga 9800164	E17 jan/20	Baixo Guandu	2020-I-22	Pebble	Trancoso, M.S.
BRDE00A15	VIÉS Microalga 9800165	E0 jan/20	Colatina	2020-I-22	<i>Paspalum pilosum</i>	Trancoso, M.S.
BRDE21A15	VIÉS Microalga 9800166	E21 jan/20	Linhares	2020-I-21	<i>Panicum aquaticum</i> ; Bambusoideae	Trancoso, M.S.
BRDE22A15	VIÉS Microalga 9800167	E22 jan/20	Linhares	2020-I-21	<i>Cuphea melvilla</i>	Trancoso, M.S.
BRDE26A15	VIÉS Microalga 9800168	E26 jan/20	Linhares	2020-I-20	<i>Talipariti pernambucense</i> ; <i>Panicum aquaticum</i>	Trancoso, M.S.
BRDE18A15	VIÉS Microalga 9800169	E18 jan/20	Colatina	2020-I-22	<i>Eleocharis interstincta</i>	Trancoso, M.S.
BRDE19A15	VIÉS Microalga 9800170	E19 jan/20	Linhares	2020-I-23	<i>Eichhornia azurea</i>	Trancoso, M.S.
BRDE20A15	VIÉS Microalga 9800171	E20 jan/20	Linhares	2020-I-23	<i>Panicum aquaticum</i>	Trancoso, M.S.
BRDE24A15	VIÉS Microalga 9800172	E24 jan/20	Linhares	2020-I-20	<i>Eleocharis interstincta</i>	Trancoso, M.S.
BRDE25A15	VIÉS Microalga 9800173	E25 jan/20	Linhares	2020-I-21	<i>Typha domingensis</i>	Trancoso, M.S.
BRDE25aA15	VIÉS Microalga 9800174	E25A jan/20	Linhares	2020-I-21	<i>Nymphaea caerulea</i>	Trancoso, M.S.
BRDE17A16	VIÉS Microalga 9800175	E17 fev/20	Baixo Guandu	2020-II-05	Pebble	Santana, L.M.
BRDE00A16	VIÉS Microalga 9800176	E0 fev/20	Colatina	2020-II-05	<i>Panicum dichotomiflorum</i> ; <i>Panicum repens</i>	Santana, L.M.
BRDE21A16	VIÉS Microalga 9800177	E21 fev/20	Linhares	2020-II-04	<i>Cuphea melvilla</i> ; Poaceae	Santana, L.M.
BRDE22A16	VIÉS Microalga 9800178	E22 fev/20	Linhares	2020-II-04	<i>Cuphea melvilla</i>	Santana, L.M.
BRDE26A16	VIÉS Microalga 9800179	E26 fev/20	Linhares	2020-II-03	<i>Talipariti pernambucense</i> ; <i>Panicum aquaticum</i>	Santana, L.M.
BRDE18A16	VIÉS Microalga 9800180	E18 fev/20	Colatina	2020-II-05	<i>Eleocharis interstincta</i>	Santana, L.M.
BRDE19A16	VIÉS Microalga 9800181	E19 fev/20	Linhares	2020-II-06	<i>Eichhornia azurea</i>	Santana, L.M.
BRDE20A16	VIÉS Microalga 9800182	E20 fev/20	Linhares	2020-II-06	<i>Panicum aquaticum</i>	Santana, L.M.
BRDE24A16	VIÉS Microalga 9800183	E24 fev/20	Linhares	2020-II-03	<i>Nymphaea lingulata</i>	Santana, L.M.
BRDE25A16	VIÉS Microalga 9800184	E25 fev/20	Linhares	2020-II-04	<i>Typha domingensis</i>	Santana, L.M.
BRDE25aA16	VIÉS Microalga 9800185	E25A fev/20	Linhares	2020-II-04	<i>Nymphaea caerulea</i>	Santana, L.M.

be submitted non-periodically. This series is the first effort to describe periphytic algae flora from this region.

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Authors Contributions

Stéfano Zorzal-Almeida, Karoline Barros de Souza, Lucineide Maria Santana, Bianka Gerhardt Endlich, Bruna Fadul-Souza, Davi Barbosa de Jesus, Izabela Clara Marques Balinhas, Mateus Sardi Trancoso, Miguel Ângelo Scardua-Filho, and Valéria de Oliveira Fernandes: contributed equal to the concept, design, and critical revision.

Stéfano Zorzal-Almeida: contributed to the first draft of the manuscript.

Conflicts of interest

There is no conflict of interest.

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